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SAMPLING MICROBIOLOGICAL AEROSOLS

Harold W. Wolf, et al

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Sampling Microbiological Aerosols

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I. Introduction

The science of air hygiene encompasses the assay and control of radiological, chemical, biological, and inert material contamination of air and particularly the effect of these contaminants on human health and well-being. Studies of the chemical and inert material pollution of the air have been progressing at expanding rates since the widely publicized London and Donora smog episodes, neither of which was the first such episode to occur in this age of industrialization. The monitoring of air for radiological activity has also progressed rapidly as a result of nuclear weapons testing.

Studies in the field of biological air analysis have not, until recently, had such stimulation. In the past, episodes in hospitals, nurseries, operating rooms, and biological laboratories have involved few people during isolated incidents, and the problems of acute upper respiratory illnesses have not usually been dramatic except for the influenza epidemics. However, interest in biological air contamination and the role of airborne contagion has recently been stimulated by the current wave of antibiotic resistant staphylococcal infections occurring in hospital environments (*1*), the tremendous impact on the Nation's health of minor upper respiratory illnesses as revealed by the recently inaugurated U.S. National Health Survey (*2*), and the increasing realization of the potential importance of biological weapons in warfare.

Biological contaminants in air include, in addition to bacteria, the fungi, viruses, and pollens. This monograph is concerned primarily with bacterial contaminants. However, airborne fungi and viruses can be sampled satisfactorily with several of the devices and techniques described. In general, the techniques described are intended only for numerical determinations of the bacterial flora of air. For species identification of the collected bacteria, standard bacteriological procedures employed in public health and clinical laboratories can be used.

Biological contaminants occur in the air as aerosols, which are defined as solid or liquid particles suspended in air. Particulates in a biological aerosol usually vary in size from less than 1 micron to approximately 50 microns or possibly larger. These particles may consist of a single unattached organism or may occur in the form of clumps composed of a number of bacteria. The organism may adhere to a dust particle or may exist as a free floating particle surrounded by a film of dried organic or inorganic material. Vegetative types of bacterial cells are probably present in the air in a lesser number than bacterial spores but are more important to health, since they include the primary etiological agents of communicable diseases. Many vegetative cells ordinarily will not survive very long in the air unless the relative humidity and other factors are favorable for survival of the organism and the organism is enclosed with some protective cover. Pathogens which resist drying, such as the staphylococcus, streptococcus, and the tubercle bacillus, will survive for relatively long periods, and therefore they may be carried considerable distances while still viable, or they may settle on surfaces to be again made airborne as a secondary aerosol during activities such as sweeping and bedmaking.

Prior to initiating a sampling program, the investigator should be cognizant of some of the characteristics of the aerosol and any peculiarities of the organisms in which he may be particularly interested. It must first be decided whether he wants to know the number of viable organisms suspended in a sample of air, the number of particles bearing viable organisms, the size distribution of the particles, or information relating

to all of these factors. He must consider the conditions of the aerosol to be sampled, such as the relative amount of inert material present, the environmental factors affecting the aerosol, and the probable concentration of organisms. If the purpose of the sampling program is to determine the number of respirable organisms, only those particles of 5 microns or less should be investigated, because larger particles are usually trapped either in the nasal passages or in the upper respiratory tract by droplets of saliva, by mucus, and cilia (*3*). These large particles are generally eliminated from the body. All of these items are important when determining the type of sampler to be employed, the sampling time, and the duration of the sampling program.

There are still other factors to consider before initiating an extensive air sampling program, for instance, the type of sample and sampler desired. The number of samples to be taken, and tests to be made should be determined statistically. The availability of sufficient sampling equipment and supplies to complete the program should be assured before starting the work. Arrangements for proper bacteriological examination of the collected samples should be made. If the samples cannot be assayed immediately, they must be refrigerated to prevent the growth or the loss of viability of the organisms. In determining the method of assay, the collecting and culturing media must be selected which will be most satisfactory for the growth of the organism or organisms in which one is interested.

A wide variety of instruments is available for sampling airborne bacteria. Many of these are not considered pertinent to this report, hence only those which would be most useful are discussed. Many of the samplers described have been studied by the U.S. Army Chemical Corps or Public Health Service laboratories, but only a few of these studies have been reported in the literature. The internal laboratory reports of the two organizations concerning these studies, in general, are not available for public distribution. Samplers described in the open literature are referenced in this monograph. Some of the samplers are self-contained units requiring only a power supply and the appropriate collecting medium in order to be used. The majority, however, require auxiliary equipment such as a vacuum pump and an airflow measuring device. Therefore, limited information on auxiliary equipment has been included in section VII.

II. Bacterial Air Sampling Methods

Methods for sampling airborne bacteria are basically the same as the methods used to sample dust and other airborne particulates. Since it is necessary to preserve the viability of the bacteria while not permitting growth, the existing samplers have been modified for the recovery of living biological agents. The basic methods include:

- Impingement in liquids.
- Impaction on solid surfaces.
- Filtration.
- Sedimentation.
- Centrifugation.
- Electrostatic precipitation.
- Thermal precipitation.

A brief discussion of these methods and their application to bacterial sampling follows.

Impingement in Liquids

Samplers which utilize the principle of impingement and washing of air, in which organisms are entrapped in a liquid medium, will be referred to as liquid samplers. In high velocity liquid impingers, air is drawn through a small jet and is directed against a liquid surface and the suspended particles are collected in the liquid. High velocity im-

pingers are constructed so that the air inlet tube projects into the liquid or is in close proximity to the liquid surface. The airstream approaches sonic velocity in the high velocity liquid samplers, resulting in almost complete collection of suspended particles, but this condition tends to cause the destruction of vegetative cells (*4*). For the recovery of some vegetative cells, the highest efficiency appears to occur when the jet velocity is approximately 70 percent of sonic velocity. Due to the agitation of the collected particles in the collecting liquid, bacterial clumps are likely to be broken into smaller fragments or even into unicellular particles. For this reason, the bacterial counts obtained with this type of sampler tend to reflect the number of bacterial cells suspended in the air.

In liquid samplers which utilize the air-washing principle, air enters the sampler at a low velocity through either a large jet, a fritted glass disk, or a perforated tube. The airborne particles are wetted and entrapped in the collecting medium. Since the air is bubbled through the liquid, fragmentation of bacterial clumps will occur, but this may not be as extensive as in the high velocity impingers. There is also less likelihood of destruction of the collected bacteria as compared with the high velocity liquid impingers. The bubbler or air-washing type of sampler is not efficient for the collection of small (less than 5 microns) airborne particles, since these small particles tend to remain in the air bubbles and are carried out of the sampler with the discharged air (*5*). The Shipe or tangential jet, the critical orifice liquid impinger (*6*), and the all-glass impinger (Chemical Corps type) are examples of samplers that operate by the impingement principle at an air velocity approaching sonic. The Greenburg-Smith (*7*, *8*) and M.S.A. midget impingers (*9*) operate by the same principle. However, air enters the sampler at a velocity less than that of air entering the tangential jet (*10*) or the all-glass impinger (Chemical Corps type). Samplers in which the air-washing principle is used are the Folin bubbler (*11*), which bubbles the airstream through the liquid, and the venturi scrubber (*12*), which washes the air by wetting the airborne particles with a fine liquid spray and impinges the wetted particles against a glass surface.

The selection of a collecting medium for liquid samplers is dependent upon the particular organism being isolated. The primary consideration in determining a suitable medium is the preservation of the bacteria recovered. In quantitative studies a medium must be employed which will minimize the multiplication or death of the organism. The more common collecting media include: buffered gelatin, tryptose saline, peptone water, and nutrient broth. Under certain conditions it may be desirable to use an antifoam agent in the collecting medium to prevent excessive foaming and carryover of the liquid, since this may result in the loss of entrapped bacteria. Dow-Corning Anti-Foam A and B, General Electric Anti-Foam 60, and olive oil are some antifoam agents which may be used.

Atmospheric conditions must be considered when utilizing liquid impingers for recovery of bacteria in air. If the temperature is high and the relative humidity low, rapid evaporation of the collecting liquid will result, thus limiting the length of the sampling period. If the ambient temperature is below 40° F., freezing of the collecting liquid is likely to occur. A nontoxic low freezing point diluent such as glycerol may be added, or some means of temperature control may be used, to prevent freezing during the sampling period.

In quantitative studies careful measurement of the total airflow is required in order that the number of organisms per specific volume of air can be determined. The volume of collecting fluid at the completion of a sampling interval must also be measured in order to determine the number of cells collected.

Impaction on Solid Surfaces

Impactors deposit particles suspended in the air directly on solid surfaces. The bacterial count obtained with this type of sampler reflects the number of airborne particles carrying bacteria. Certain samplers operated by this principle also have been designed

to determine the particle-size distribution of airborne micro-organisms.

If the volume of the sampled air is accurately metered, a quantitative estimate of the airborne bacteria can be determined. The solid surface on which the particles are collected may be dry, coated, or an agar surface. Samplers which collect particles on dry or coated surfaces, such as the cascade impactor (*13, 14*), are used for determining the number of particles and the size distribution by means of microscopic examination of the deposits. The number and size distribution of viable organisms then can be approximated by washing the particles from the collection surfaces and enumerating by bacteriological methods. However, only resistant organisms, such as spores, will remain viable on dry or coated surfaces for an appreciable length of time. In the surface-washing process large particles which may contain many viable organisms may be broken into individual organisms. The result would then indicate the number of viable organisms present but not the number of particles collected.

Samplers which collect particles on agar surfaces are used to determine the number of viable particles present, since bacterial colonies grow directly on the collecting surface. The sieve (*15, 16*), slit (*17–20*), Andersen (*21*), and single-stage impactor samplers, when used with an agar collecting surface, are examples of solid medium impactors. The Andersen, the cascade slit, and the single-stage impactor also can be used to determine the size distribution of the viable particles in an aerosol.

The sieve and Andersen samplers are operated by drawing air at a rate of 28.3 liters per minute through a large number of small, evenly spaced holes drilled in a metal plate. The suspended particles are impacted on an agar surface located a few millimeters below the perforated plate. The Andersen sampler consists of a series of six sieve type samplers which have holes of smaller diameter in each succeeding plate. The velocity of the incoming air increases for each succeeding plate, resulting in a separation of the particles into six size ranges. From the number of colonies growing on each agar plate, the particle count as well as the particle-size distribution can be determined. In the operation of slit samplers, air is drawn through a narrow inlet slit, and particles are impacted on an agar surface contained in a moving dish or tray. The slit samplers permit determination of a time and viable particle concentration relationship. Samples from the sieve, slit, and Andersen type samplers are assayed by incubating the exposed plates and counting the developed colonies.

These samplers require a vacuum source sufficient to draw a constant flow of air through the sampler—usually 28.3 liters per minute, although the airflow through the slit sampler may be changed by altering the dimensions of the slit.

In the operation of a single-stage impactor, the air is drawn through a small orifice at a constant airflow of 12.5 liters per minute, which impacts the larger particles on the surface of a small dish of agar or on a glass disk. The smaller particles are carried on through the sampler and are collected in a suitable final collector such as a filter or liquid impinger. By changing the diameter of the inlet orifice, the size range of particles collected on the agar or glass disk may be changed. Results of samples collected indicate the number of viable organisms in particles of a size range that are above and below the cutoff point of the sampler.

The cascade impactor consists of a series of four or five stages; each stage has a small air inlet slit which causes the particles to be impacted against a glass slide. In each successive stage, the slit area decreases, resulting in a higher velocity of the passing airstream, so that a separation of particles into size ranges results. The addition of a thin layer of adhesive to the slides aids the collection of particles. Long sampling periods may cause desiccation of certain organisms and overcrowding of particles so that microscopic sizing is difficult. The total number of viable organisms can be determined by washing the particles from each of the slide samples into sterile water and assaying by standard bacteriological techniques.

Filtration

In the filtration of bacterial aerosols, the particles either strike the fibers of the filter directly or are attracted to them and held in place by electrostatic and other forces. The particles are not normally removed from the airstream by a simple screening or sieving action, except in the case of membrane filters (*22–27*). An efficient filter is usually composed of fibers, generally perpendicular to the direction of the airflow and having a diameter equal to or less than the diameter of the particles collected. The mesh or opening between the fibers may be quite large compared to the size of the particle (*28*). Filtration efficiency will increase if the filter thickness or number of fibers is increased, but the resistance to airflow also will increase.

Numerous materials are used as filter media (*29–35*). Some of these materials are cellulose-asbestos paper, glass wool (*36, 37*), cotton (*38*), alginate wool, gelatin foam (*39, 40*), and membrane filters.

Membrane filters differ from the other types of filters in that they mainly sieve the particles from an aerosol. The pores of the membrane are smaller than the diameter of the collected particles and therefore the particles do not penetrate into the filter but are retained on or within a few microns of the filter surface.

When assaying most filter samples, the complete filter or a section of it is agitated in a suitable liquid until the particles are uniformly dispersed. Aliquots of the suspension are then assayed by appropriate bacteriological techniques. The bacterial counts obtained are usually higher than the actual number of viable particles in the aerosol, since clumps of bacteria may be fragmented during agitation of the sample.

The viability of some organisms is detrimentally affected if the bacteria remain on the filter during long sampling periods, because the continuous passage of air over organisms will result in desiccation. This effect is reduced at high relative humidities.

Table 1. Settling rates of airborne particles

Diameter of particles (microns)	Velocity of settling	
	Feet per minute	Inches per hour
0.1	0.00016	0.115
2	.00036	.259
4	.0013	.936
6	.002	1.44
8	.005	3.60
10	.007	5.04
20	.024	17.3
40	.095	68.4
60	0.21	
80	0.38	
100	0.59	
200	2.4	
400	9.5	
600	21.3	
800	37.9	
1000	59.2	
2000	352	
4000	498	

NOTE: This table has been compiled from "Size and Characteristics of Air-Borne Solids," by W. G. Frank, published in the Smithsonian Meteorological Tables. Rates are for particles in the shape of spheres, having a specific gravity of 1.0 and settling in air at a temperature of 70° F.

Sedimentation

A simple method of sampling airborne particulates can be accomplished by the use of the sedimentation process (*17, 41*), whereby particulates suspended in the air are allowed to settle on plain surfaces or on surfaces coated with a nutrient medium. Application of this method can be used to determine the number of viable particles, the total number of viable organisms, and the total number and size of all particles that settle in a given time. This method of sampling is obviously more suitable for the enumeration of larger sized particles, since they settle more rapidly than the smaller sized particles. Table 1 gives the settling velocities of various sizes of particles of unit density in still air.

Samples collected by sedimentation provide only a qualitative index of the particles suspended in the air. Air movement will influence the deposition of the particles so that the particle-size distribution may indicate a higher percentage of large particles than is actually present in the air. When an aerosol is collected in a closed container of known volume and the particles are allowed to settle quiescently onto a collecting surface, a

quantitative measure of the number of particles suspended in the air can be obtained bacteriologically and a true particle-size distribution can be obtained by microscopic examination.

The number of viable particles that settle from the air in a given time is determined by allowing the particles to settle on a nutrient surface such as an open agar Petri dish or agar coated slide, and the undisturbed particles are allowed to develop into visible colonies during subsequent incubation. The number of settled particles is then determined from the colony count.

The total number of viable organisms settling from the air in a given time is determined by allowing the particles to settle on surfaces from which they can be removed by washing in a suitable liquid. The collecting surface is agitated thoroughly in the wash liquid to break up clusters of organisms and to dislodge the bacteria from inert particulate matter, so that each individual cell may develop into a separate colony when the liquid is plated on a nutrient medium.

The number and the size of all particles settling from the air in a given time are determined by allowing the particles to settle on glass slides, membrane filters, or other surfaces. The particles are then counted and sized microscopically without removal from the surface on which they have been collected.

Advantages of sedimentation samplers are: inexpensiveness, simplicity of use, and ability to collect the particles in their original state. The disadvantages are: inability to give quantitative counts unless the aerosol sample is allowed to settle in a closed container, a long waiting period is required for all particles to settle, and some loss in viability may occur during the settling process.

Centrifugation

Since deposition of particles by sedimentation in the size range of bacteria is not always satisfactory or efficient, a few bacterial samplers are available which employ centrifugal force for propulsion of the particles to the collecting surface. When the aerosol moves in a circular path at high velocity, the suspended particles are impacted on the collecting surface by a force proportional to the particle velocity and mass. The particles are usually collected on glass or agar surfaces, which are removed from the sampler and assayed by appropriate methods. Two samplers of this type in use are the cyclone (aerotec tube) and the air centrifuge (Wells) (*42–44*). In the cyclone type, the sampler remains stationary while the aerosol travels in a circular path, and the large particles are collected at the bottom of the sampler. In the air centrifuge type sampler, the collecting vessel and aerosol rotate at high velocity, resulting in the impaction of particles on the walls.

Electrostatic Precipitation

Samplers using this method collect particles by drawing a given amount of air at a measured rate (up to 85 liters per minute) over an electrically charged surface (*45–49*). In some samplers the air is ionized prior to passing over the collection surface. The electrostatically charged particles are subsequently collected on either positively or negatively charged surfaces. These samplers may employ a variety of solid collecting surfaces such as agar or glass. Although these samplers have a relatively high sampling rate, high collection efficiency, and low resistance to airflow, they are complex and must be handled carefully. Furthermore, little is known about the effect on viability and clumping of electrostatically charged particles.

Thermal Precipitation

Samplers which collect particles on surfaces by means of thermal gradients are classified as thermal precipitation samplers. Their design is based on the principle that airborne particles are repelled by hot surfaces and are deposited on colder surfaces by forces proportional to the temperature gradient. These samplers are used for determination of particle-size distribution and are more effective when collecting small particles (1 micron and less). Several samplers of this type have been described in the litera-

ture (*50–52*). However, they are not commonly used since they require careful adjustment and the air sampling rates are quite low (3–400 ml. min.).

The particles are usually collected on a glass coverslip or electron microscope grid, and the particles are subsequently sized and counted microscopically.

III. Particle Sizing

Consideration should be given to the determination of particle size when studying airborne micro-organisms and their relation to respiratory infections, since the amount of particulate material and bacteria retained in the respiratory system is largely dependent upon the size of the inhaled particles. Considerable information is available concerning the selective action of the respiratory system for various sized particles. Hatch and others (*53–57*), have found that particles larger than 5 microns are efficiently trapped in the upper respiratory tract and are removed by ciliary action or some other means. Particles in the 1- to 5-micron range which reach the alveoli are usually retained. However, retention decreases for particles from 1 micron down to 0.1–0.2 micron. Recent evidence indicates that the alveolar retention again increases for particles less than 0.1 micron in size.

The following brief discussion is presented to acquaint the reader with a common method of particle-size determination of airborne micro-organisms. Numerous texts (*58*) and articles (*59,60*) devoted to particle sizing are available if a more detailed and theoretical discussion is desired.

Particles aerosolized from clean suspensions of micro-organisms are usually spherical in shape. However, bacteria found in the day-to-day atmosphere are generally associated with foreign material and may be present as particles of irregular shapes. When particles are spherical, their size can be reported in terms of diameter or some other related dimension. However, if the particles are not spherical, size can be reported in terms of length, width, volume, or mass. To facilitate reporting the size of airborne bacterial particles, it can usually be assumed that all particles in a given sample have the same density and shape. To simplify further the determination of bacterial particle size, it can be assumed that all the bacterial particles are spherical and that the mean diameter need only be approximated. Although this is not precise, it will provide the necessary information for all practical purposes.

The method most often used for size analysis (*61, 62*) of bacterial particles is direct measurement with a light microscope and a calibrated scale or graticule. The particles are collected without disintegration on a glass slide or on a membrane filter so that there are between 25 and 150 particles per field. A total of 200–500 particles should be counted and sized so that a representative sample of the deposited particles is obtained. The most useful graph to show size variation in particle sizing work is the cumulative type, in which the cumulative percent count, or mass less than a stated size, is plotted against that size on logarithmic probability paper. The count median diameter, mass median diameter, and geometric standard deviation are determined from this graph. An example of these determinations from hypothetical data follows.

After 200 particles from the sample slide are counted, sized, and tabulated (table 2), the percent of the total number is calculated for each size and these percentages are then cumulated for determination of count median diameter. If the mass median diameter is desired, the total mass for each size group is calculated by cubing the diameter and multiplying by the number of particles in that size group. Since the density and the

Table 2. Particle-sizing data

Diameter (microns)	Number counted	Percent of total	Cumulative percent	Volume (diameter ^{3} x number)	Percent of volume	Cumulative percent by volume
1	5	2.5	2.5	5	0.01	0.01
1.4	14	7	9.5	38	.10	.11
1.9	23	11.5	21	157	.43	.54
2.7	41	20.5	41.5	807	2.19	2.73
3.8	57	28.5	70	3,125	8.50	11.23
5.4	29	14.5	84.5	4,566	12.41	23.64
7.7	20	10	94.5	8,130	21.82	48.46
10.9	9	4.5	99	11,655	31.68	80.14
15.4	2	1.0	100	7,304	19.85	99.99
Totals	200	100		36,789		

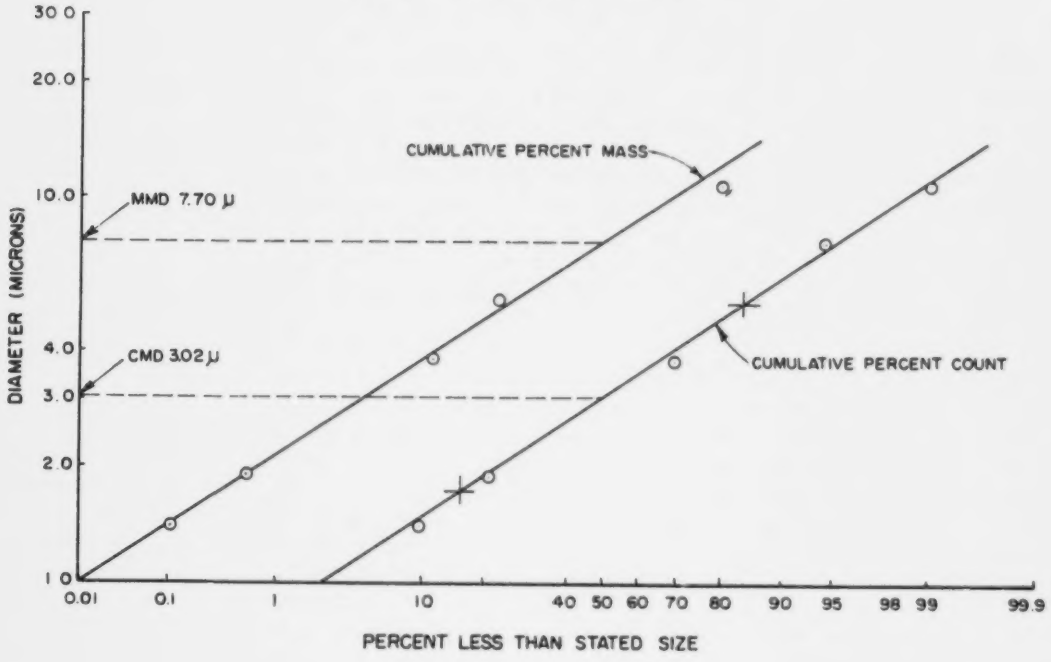
factor $1/3\ \pi$ are constant, they are eliminated from the calculation. The percent of the total mass is calculated for each size group, and these percentages are then cumulated for determination of mass median diameter. The cumulated figures for number percent and mass percent are plotted against the corresponding size as shown in figure 1. By reading the size corresponding to the 50 percent intercept as shown on the graph, the count median diameter (CMD) is found to be 3.02 microns and the mass median diameter (MMD), 7.70 microns.

The geometric standard deviation is found by substituting values from the graph in the following relationship (*63*):

$$\sigma_g = \frac{84.13 \text{ percent size}}{50 \text{ percent size}} = \frac{50 \text{ percent size}}{15.87 \text{ percent size}} = 1.7$$

The amount of work involved in sizing an aerosol can be reduced considerably by using

Figure 1. Particle-size distribution



such samplers as the Andersen, TDL slit, and cascade impactor, which separate the particles into size ranges as the sample is collected. However, these samplers should first be calibrated by sampling similar aerosols under the same conditions as those for which they are to be used. Particles collected on each stage should be measured microscopically to determine CMD or MMD as explained previously. When the instrument has been calibrated, it can be assumed that the CMD or MMD for each stage will remain relatively constant, providing the particulate material and sampling conditions do not change.

IV. Selection of Samplers

Many types of air samplers and modifications thereof have been described in the literature, each of which has been designed for individual needs and specific conditions or redesigned in an attempt to improve its efficiency. There is still a need for improvement of existing bacterial air samplers, since the collection of airborne bacteria, without changing such properties as size and viability, is difficult to accomplish.

When selecting a sampler for a specific air sampling program, many factors must be considered, but first one must have a clear understanding of the type of information desired and the particular determinations that must be made. For instance, is information desired on (*a*) one particular organism or all organisms that may be present in the air, (*b*) the concentration of viable particles or of viable organisms, (*c*) the change of concentration with time, (*d*) the size distribution of the collected particles, or (*e*) a combination of these points? Furthermore, are the results on concentrations to be qualitative or quantitative?

Factors to be considered before selecting a particular sampler are:

1. Viability and type of the organism to be sampled.
2. Sensitivity of particles to sampling.
3. Assumed concentrations and particle size.
4. Volume of air to be sampled and length of time sampler is to be continuously operated.
5. Background contamination.
6. Ambient conditions.
7. Sampler collection efficiency.
8. Effort and skill required to operate sampler.
9. Availability and cost of sampler.
10. Availability of auxiliary equipment and facilities, such as vacuum pumps, electricity, and water.

When selecting a sampler for the collection of airborne bacteria, it must be kept in mind that although the bacteria may be present as individual organisms, they generally occur as clumps or mixed with or adhering to particles of foreign matter, such as dust and organic substances. Complete or partial disintegration of these clumps will usually occur when certain types of samplers (particularly those using liquid media) are used. The results would then indicate a greater number of viable clumps or particles than was actually present in the air at the time of sampling. When the clumps or particles are completely disintegrated, the result will indicate the total number of individual organisms present in the air. When reporting the bacterial concentrations determined by various air samplers, the results should indicate whether the concentrations represent individual organisms or particles bearing bacteria.

The air sampling devices described in this monograph can be placed in the following categories according to the purpose for which they are used:

Determination of the concentration of viable particles.

Determination of the concentration of viable organisms.

Determination of the change of concentration with time.

Determination of the size of the collected particles.

Concentration of Viable Particles

Samplers used for this purpose should deposit the particles (without disintegration) on the surface of a solid nutrient medium on which the particles can grow or multiply into discrete colonies with suitable incubation. If a qualitative estimation of the number of viable particles present in the air is desired, an open agar plate may be used. If quantitation is desired, the slit (*19*) or sieve (*15*) sampler may be employed, since these samplers permit the viable particles in a measured volume of air to be deposited uniformly on an agar surface. All samplers employing agar as a collecting surface are limited to use in temperatures above 32° F. unless some method of heating is provided.

Membrane filters can be used to collect viable particles at temperatures below freezing. However, this method of sampling is limited to the collection of micro-organisms which are resistant to drying.

Concentration of Viable Organisms

A general requirement for the determination of the number of viable organisms present in the air is that the collected particles must be disintegrated during the sampling process or in the subsequent assessment of the collecting medium so that individual organisms, when placed in the proper growth medium, can develop into discrete colonies. Samplers generally used for this purpose are the liquid impingers (*64*) since they are very efficient in the collection of all micro-organisms. The sampling time of these samplers is limited to relatively short periods because of the evaporation of the collecting fluid and the killing effect upon the organism. Their use is generally limited to temperatures above 40° F. unless some method of heating is applied or a suitable antifreeze agent is added to the collecting fluid.

Samplers employing filter media and non-nutrient solid impaction surfaces can be used to determine the concentration of viable organisms in the air. However, these samplers are limited to the collection of spores and other microbial forms which are resistant to the drying usually associated with this method of sampling.

Change of Concentration With Time

The variation of concentration of viable particles or organisms with time can be determined by collecting intermittent samples with any suitable air sampling device. A simpler method of obtaining this information is by the use of automatic samplers such as slit samplers that collect continuous samples on a rotating agar surface, the modified filter tape sampler (*33*) which collects bacterial spore samples automatically on filter media, and the liquid samplers (*65*) which collect samples in a series of bubblers.

Size of the Collected Particles

The Johnson single-stage impactor, the pre-impinger (*66*), and the Casella cascade impactor (*67*) are devices which can be used to separate all collected particles into either two or four general size ranges. By direct microscopic examination of the collecting surfaces, it is possible to measure the size of the collected particles. When the collecting surfaces are biologically assayed by washing in a suitable liquid, the results represent the total number of viable organisms associated with the particles recovered on the various stages of these samplers.

The TDL cascaded slit and the Andersen samplers are two devices which can be used to separate all collected viable particles into four and six general size ranges, respectively, by direct impaction on agar media. The plate counts represent the number of viable particles containing vegetative and spore forming micro-organisms.

The final selection of an air sampling device for use in specific air sampling problems must be left to the judgment of the investigator. Consideration should be given to the factors discussed in this section and to the

individual characteristics of the candidate samplers.

No one type of sampler and assay procedure exists which can be used to collect and enumerate 100 percent of the airborne organisms. It has been the experience of the writers that the most satisfactory samplers are the sieve (limited to low concentrations), slit, all-glass impinger, and the tangential jet impinger. In any case, the sampler chosen should have an adequate sampling rate in order to collect a sufficient number of particles within a reasonable time period so that a representative sample of air is obtained for biological analysis.

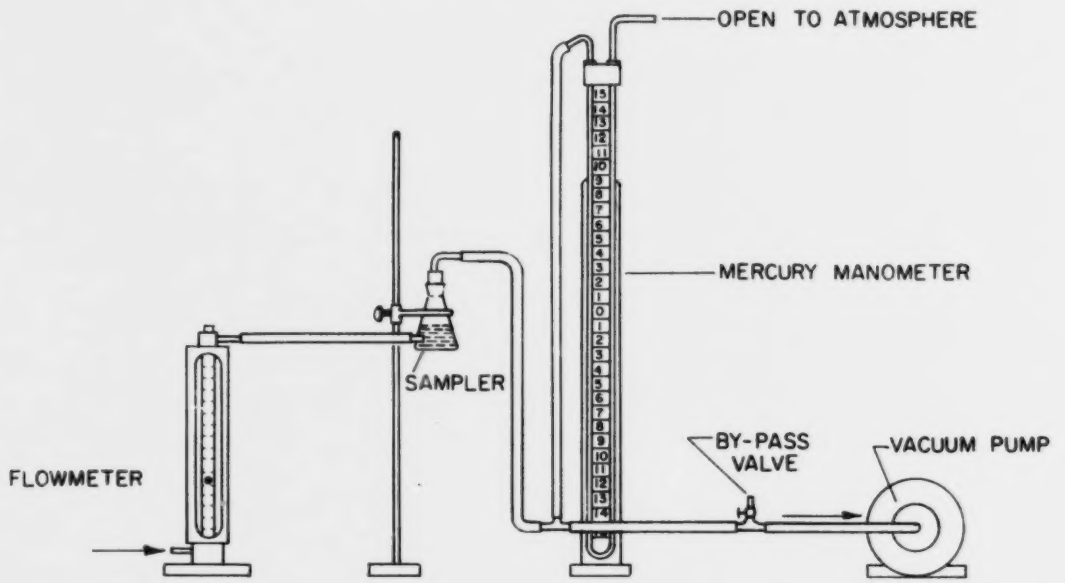
Samplers listed in tables 3–8 are more thoroughly described and illustrated in section VII, pages 18 to 50. Figure numbers in these tables refer to this more detailed description.

V. Operational Methods

Control of Air Sampling Rate

If sampling is to be quantitative in nature, the amount of air passing through the samplers must be known. The rate of flow for each sampler may be determined prior to the utilization of the device for collecting air samples. Many devices for metering the flow are available. However, three devices are described herein. The simplest but most expensive method is the use of a calibrated flowmeter connected in the vacuum line between the sampler and vacuum source. A somewhat less expensive method is the use of a manometer or a vacuum gauge and a regulating valve. To utilize this method, a calibrated flowmeter is first connected to the inlet of the sampler for calibrating the manometer. One side of a U-tube manometer or vacuum gauge is connected to the vacuum line between the sampler and the source of vacuum. The other side of the manometer is open to the atmosphere. With this system

Figure 2. Calibration of airflow



in operation, the amount of air passing through the sampler for a particular manometer reading is determined from the flowmeter and regulated by a bypass valve, preferably of the needle type, located on the vacuum inlet to the pump. In subsequent utilization of the sampler for collecting airborne micro-organisms, the flowmeter is not included in the system. The rate of flow is maintained by adjusting the vacuum to the same manometer reading recorded for the desired flow rate. This calibration will be accurate as long as the resistance of the sampler does not change (fig. 2).

The third method of metering airflow, which eliminates the need of individual flowmeters for samplers when a number are in use, is the insertion of a capillary tube in the vacuum line. This tube may be used as a critical orifice to keep the sampler flow at the desired rate. This is based on the fact that the velocity of a gas flowing through an orifice will reach a maximum or acoustical velocity when the critical pressure is reached. The theory applies to orifices in a thin flat surface, but for all practical purposes it is also applicable to short lengths of capillary tubing. The critical pressure ratio is represented by the ratio of the downstream absolute pressure to the upstream absolute pressure. This critical ratio is approximately 0.5. When a vacuum is applied to the downstream side, the flow through the orifice will increase until this pressure ratio is reached. Increasing the vacuum further will not increase the flow no matter how high it may go, provided the upstream conditions do not change (fig. 3).

This means that constant flow can be maintained through a critical orifice without the necessity of having a constant vacuum as long as one uses a vacuum source of sufficient capacity so that the downstream pressure is always less than half the upstream pressure. How much less than 0.5 this ratio is does not matter.

An orifice may be cut from a glass or metal capillary tube in a convenient length (2 inches). The outside diameter should be large enough to fit snugly inside the vacuum

Figure 3. Critical airflow through orifices

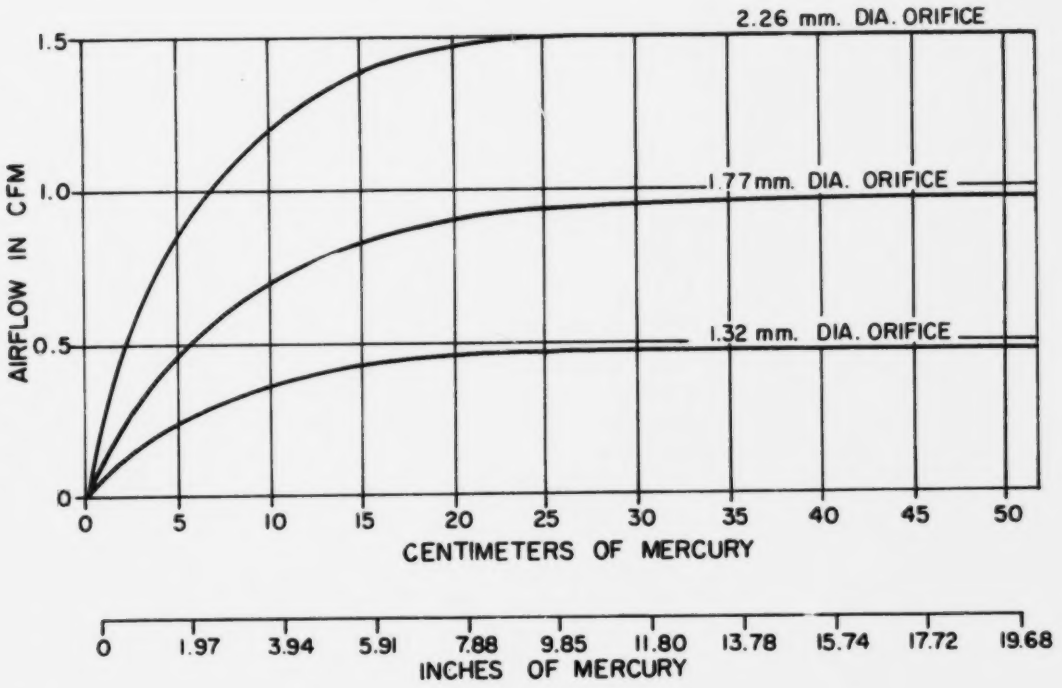
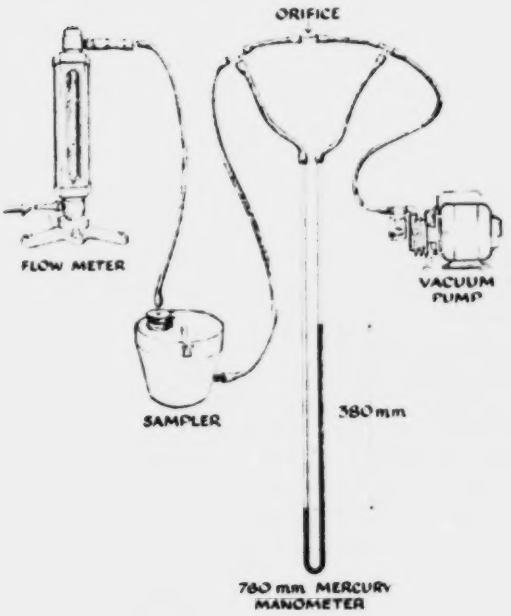


Figure 1. System for calibrating a critical orifice



tubing, and the inside diameter should be of a size to give approximately the desired flow. The exact flow through this orifice must be accurately measured, taking care to do this in the same arrangement in which it will be used so that there will be no change in conditions when samples are taken. To measure or calibrate the orifice, proceed as follows: Place the orifice in the vacuum line between the sampler and the vacuum source, and connect a standard flowmeter to the inlet of the sampler to measure the sampling rate accurately. Connect a mercury manometer across the orifice and adjust the vacuum so that the pressure differential is 38.0 cm. of Hg. or above. The airflow is indicated on the standard flowmeter, and if this value is too high or too low, repeat this procedure using another orifice with a different diameter until one is found giving the desired flow. During each measurement, make certain that the pressure drop is at least one-half atmosphere. After the proper orifice diameter has been determined, more orifices of the same length may be cut from the same tube, but each of these should be calibrated separately since the capillary diameter is often not constant through the length of the same tube. If a glass capillary is used and the diameter is larger than desired, it may be reduced by holding one end in a burner flame until the opening at the end is constricted. The flowmeter is not needed after the orifices have been calibrated (fig. 4).

Sterilization of Samplers

Most bacteriological samplers should be sterilized prior to use. Sieve samplers and filtration type samplers which have been wrapped in kraft paper, and liquid impingers with cotton plugs inserted in the intake and exhaust ports, can be conveniently autoclaved. The sampling chamber of the Casella slit sampler can be removed and autoclaved, but in actual use, it will be found that it is much more convenient and adequate simply to swab the interior with a suitable disinfectant prior to each sampling period. Gaseous sterilization techniques (*68*), if available, can be used to sterilize all of the samplers.

Media

Seven air sampling methods are described in this monograph. Application of most of these methods is based upon collection either on solid media or in a liquid. The use of proper collecting and culturing media is of fundamental importance in biological air sampling, because the determination of the number of viable micro-organisms contained in the sample depends primarily on the enumeration of bacterial colonies developing either directly on the nutrient collecting media or in subsequent plate cultures of a sample aliquot.

Many collecting and culturing media are available for biological air sampling. The selection of a particular nutrient medium will depend primarily on the nutritional requirements of the organism or organisms under study and, to a lesser extent, on the type of information desired from the study, the sampling method, and the sampling conditions. When initial collection is in a liquid

medium, the micro-organisms must remain viable without growth or decay until aliquots are taken for culture. Some of the more common liquid media used are: tryptose saline, buffered gelatin, peptone water, buffered gelatin enriched with brain-heart infusion, buffered saline, and buffered water. These media are used also as diluting fluids to obtain suspensions suitable for plating. Buffered saline and buffered water are used only for the collection of spores and other resistant microbial forms.

When collection is directly on a solid nutrient medium (*69*), it is essential that a sufficient concentration of agar be used to produce a stable medium capable of withstanding the impingement action from a rapidly flowing airstream. Solid media containing an agar concentration of 1.5–2.0 percent possess adequate stability for use with all types of air sampling devices. Some of the more common solid nutrient media employed for general bacterial air sampling are: blood agar, tryptose agar, trypticase soy agar, proteose extract agar, and nutrient agar. These media are also employed for culturing the liquid collecting media by using surface plating methods or the conventional pour-plate method, and for culturing membrane filters by placing the filter directly on the agar surface or by placing the filter on blotter pads saturated with the broth form of these media.

Under certain sampling conditions, it is sometimes beneficial to incorporate into the medium chemical agents which are inhibitory to nonsignificant spreaders and other contaminants which interfere with accurate bacterial counts and the isolation of pure bacterial colonies. Some of the inhibitory agents commonly used are: crystal violet and brilliant green to suppress Gram-positive organisms, potassium tellurite to suppress Gram-negative organisms, and cycloheximide (Actidione) to suppress molds and fungi. The incorporation of inhibitory chemicals into media should not be used as routine procedure until they have been tested with a low concentration of test organisms or with the particular organism under study.

The listed media as well as other media can be prepared from either the dehydrated products supplied by biological media suppliers, or from formulas appearing in various textbooks and manuals in the field of microbiology.

The following formulas for media are in grams per liter of distilled water:

Liquid Media

Tryptose Saline

Bacto-tryptose	1
Sodium chloride	5

Adjust pH to 7.0±0.2.

Peptone Water

Bacto-peptone	20
Sodium chloride	5

Adjust pH to 7.0±0.2.

Buffered Saline

Sodium chloride	8.5
Disodium phosphate (anhydrous)	5.8
Potassium dihydrogen phosphate (anhydrous)	3.5

Adjust pH to 7.0±0.2.

Buffered Gelatin

Bacto-gelatin	2
Disodium phosphate (anhydrous)	4

Adjust pH to 7.0±0.2.

Enriched Buffered Gelatin

Bacto-gelatin	2
Disodium phosphate (anhydrous)	4
Brain-heart infusion	37

Adjust pH to 7.0±0.2.

Buffered Water (Stock)

Potassium dihydrogen phosphate	34

Adjust pH to 7.2. Add 1 to 2 ml. of stock to 1 liter of distilled water.

Solid Media

Blood Agar Base

Beef heart, infusion from	500
Bacto-tryptose	10
Sodium chloride	5
Agar	15

Adjust pH to 7.0±0.2. Plus 5 percent fresh defibrinated rabbit, sheep, or beef blood.

Trypticase Soy Agar

BBL-Trypticase	15
BBL-Phytone	5
Sodium chloride	5
Agar	15

Adjust pH to 7.0±0.2.

Fortified Nutrient Agar

Bacto-beef extract	3
Bacto-peptone	10

Yeast extract	5
Dextrose	10
Agar	20
Adjust pH to 7.0±0.2.	
Tryptone Agar	
Bacto-tryptose	20
Dextrose	1
Sodium chloride	5
Agar	15
Thiamine hydrochloride	0.005
Adjust pH to 7.0±0.2.	
Proteose Extract Agar	
Proteose peptone (Difco)	20
Bacto-beef extract	3
Bacto-yeast extract	3
Malt extract (Difco)	3
Dextrose	5
Sodium chloride	8.5
Agar	20
Adjust pH to 7.0±0.2.	

Processing Samples

Samples should be processed as soon as possible after collection. Unless liquid samples can be processed shortly after being exposed, they should be kept at low temperatures, since variations may occur in the results due to growth or death of the entrapped organisms prior to plating. Long intervals of time between collection and incubation may also be detrimental to the organisms on agar plates. The most desirable situation would be to provide the necessary facilities at the location where samples are being collected. This can be accomplished by using a mobile laboratory unit at the sampling station.

Computation of Sampling Data

The problem of monitoring the levels of airborne micro-organisms requires the recording of a substantial volume of pertinent information from a variety of sources. A comprehensive analysis of the data is difficult unless a systematic method of tabulating is available. One method of recording data which has been found satisfactory in previous studies is shown in figure 5. Forms I and II are used at the sampling site, and Form III in the laboratory.

Assuming that each colony on the agar plate is the growth from a single bacteria-carrying particle, the contamination of the air in question is determined from the number of colonies counted. The airborne micro-organisms may be reported in terms of the number per cubic foot of air sampled.

The following formulas can be applied to convert the colony count to organisms per cubic foot of air sampled:

For solid agar samplers

$$\frac{C}{R \times P} = N$$

Where C = Total plate count.
R = Airflow rate, c.f.m.
P = Duration of sampling period (minutes).
N = Number of organisms collected per cubic foot of air sampled.

For liquid impingers

$$\frac{C \times V}{Q \times P \times R} = N$$

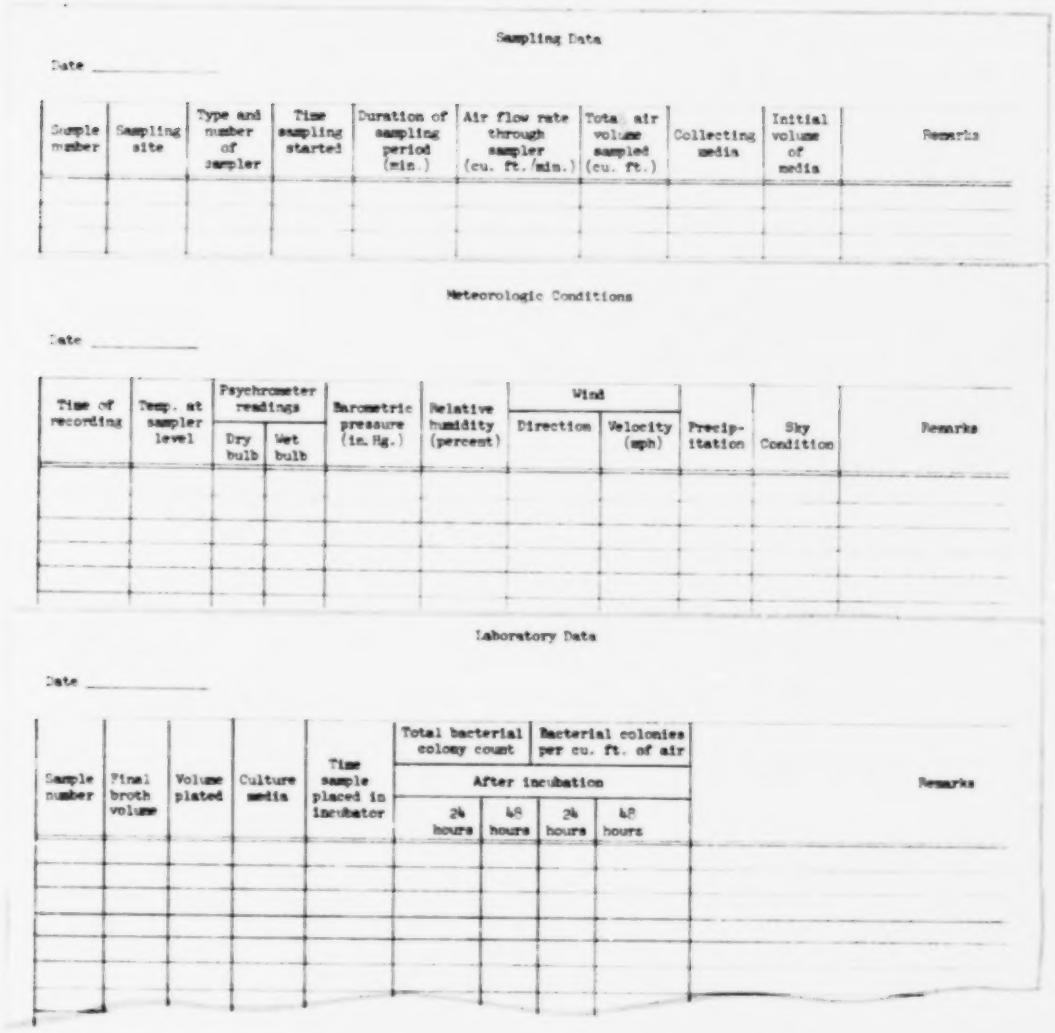
Where C = Total number of colonies from all aliquots plated.
V = Final volume in mls. of collecting media.
Q = Total number of mls. plated.
P, R, and N as above.

Planning a Sampling Program

When plans are being made for conducting an air sampling program, many obvious factors are easily overlooked that may cause delays unless proper allowances are made. The points listed below are obvious and are given only to serve as a checklist. After the type of sampler or samplers is chosen for the work as discussed in section IV, the following checklist should be considered:

1. Are enough samplers available to complete the proposed program?
2. Is the vacuum source sufficient for operation of the samplers?
3. If pumps are used, is the powerline of sufficient capacity to carry the total current load?
4. Are all pumps in good operating condition? Is there sufficient and proper lubricant available?
5. Are tools available for emergency repairs?
6. Are personnel instructed properly on the operation of samplers?

Figure 5. Sampling data forms I, II, and III



Sampling Data

Date _____

Sample number	Sampling site	Type and number of sampler	Time sampling started	Duration of sampling period (min.)	Air flow rate through sampler (cu. ft./min.)	Total air volume sampled (cu. ft.)	Collecting media	Initial volume of media	Remarks

Meteorologic Conditions

Date _____

Time of recording	Temp. at sampler level	Psychrometer readings		Barometric pressure (in. Hg.)	Relative humidity (percent)	Wind		Precipitation	Sky Condition	Remarks
		Dry bulb	Wet bulb			Direction	Velocity (mph)			

Laboratory Data

Date _____

Sample number	Final broth volume	Volume plated	Culture media	Time sample placed in incubator	Total bacterial colony count		Bacterial colonies per cu. ft. of air		Remarks
					After incubation				
					24 hours	48 hours	24 hours	48 hours	

7. Will enough sterile media or samplers be available when needed?

8. Are facilities available for storage (refrigeration) of samples until they can be assayed?

9. Are complete laboratory facilities available for assessment of samples?

10. Are proper culture media prepared for assessment of samples?

11. Is all assay equipment sterile and ready for use?

12. Are duplicate supplies of materials on hand for items easily broken or consumed, such as fuses, glassware, thermometers?

13. Have laboratory personnel been instructed in the proper methods of assay?

14. Are personnel prepared to record all data relative to sampling and assaying?

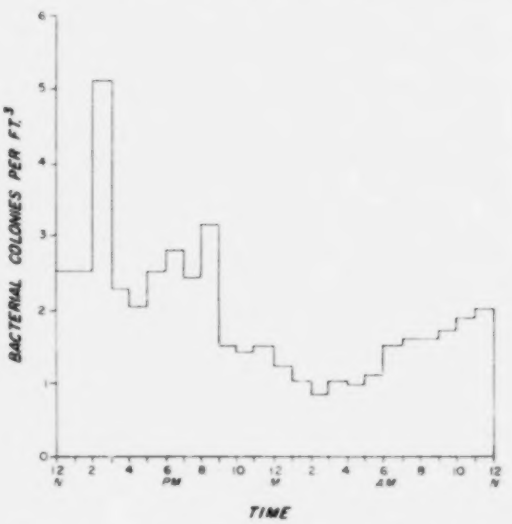
VI. Normal Bacterial Levels

Precise airborne bacterial concentrations likely to be found in various environmental situations cannot be predicted. The examples which follow are presented only as a guide.

An example of daily mean counts obtained in a highly industrialized area in the Midwest is shown in figure 6. Air samples collected during the winter months indicated a low concentration of airborne bacterial particulates. In the spring and early summer months a markedly higher concentration of airborne bacteria was observed. Although not shown in the figure, contamination of the air during the summer months was at a lower level than during the spring months, but was higher than that recorded for the winter season.

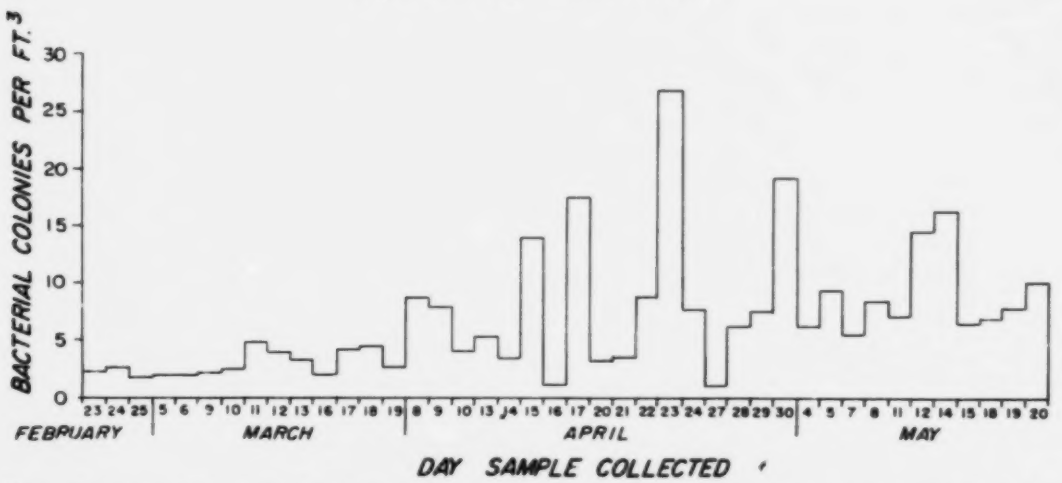
Aside from seasonal and daily differences in airborne bacterial counts, diurnal variations also occur. An example of hourly changes in counts for air samples collected in an open field of a nonurban area is shown in figure 7. Under the conditions existing

Figure 7. Hourly bacterial counts



NOTE: Extramural, sieve sample, heart infusion agar with 5 percent blood added, 37° C. incubation for 24 hours, June 27–July 3, 1951, Oatland Island, Savannah, Ga.

Figure 6. Daily bacterial counts



NOTE: Extramural, sieve sampler, heart infusion agar with 5 percent blood added, 37° C. incubation for 48 hours, February-May 1953, Detroit, Mich.

at the time, a decrease in counts was recorded between the hours of 1:00 a.m. and 6:00 a.m. and a high range from 2:00 p.m. to 8:00 p.m. It must be emphasized that the diurnal trends observed in this sample do not necessarily apply to other areas.

Indoors, the levels of bacterial contaminants are usually higher than those encountered outdoors. Concentrations depend on such factors as ventilation rates, crowding, and the degree of activity of individuals occupying the enclosure. Various workers in the field have found a wide range of counts. Under certain conditions, bacterial levels as high as 3,400 bacterial particles per cubic foot of air have been recorded in army barracks during sweeping and bedmaking activities (*70*).

Frequently some of the air within industrial plants contains pathogenic bacteria. Air samples collected at a goat-hair processing mill have shown that particles bearing *Bacillus anthracis* were present in an average concentration of 9 per cubic foot of air (*71*). Other investigators have found that airborne infectious organisms which cause Q fever in man (*72*) remain viable for considerable periods and may be transmitted from one area of a building to another. Some laboratories have reported that cross-contamination of experimental animals (*73*) has occurred by airborne transmission of bacteria. As in outdoor sampling, the levels of bacterial contaminants vary from site to site (*74*) and precise airborne bacterial levels cannot be given. It is rather safe to assume, though, that there is a greater likelihood of recovering airborne pathogens indoors than outdoors.

VII. Description of Samplers and Equipment

Table 3. Liquid impingement samplers

Common characteristics: Require use of an antifoam agent. Not well adapted to low concentrations. Size of air sample can be varied within wide limits. Sampling time dependent upon rate of evaporation of collecting liquid. Disintegration of particles can occur in various degrees. High efficiency for particles greater than 1 micron at high jet velocities. Some killing effect associated with high impingement velocities and extended continuous sampling. Quantitation by diluting and plating procedures. Final assay results expressed as organisms per unit volume of air. Generally limited to use in temperatures above 40° F. unless heated or suitable antifreeze agent is used. Easily sterilized by autoclaving. Simple to operate, portable, easily broken.

Sampler	Figure number	Sampling rate (lpm)	Volume of collecting liquid (ml.)	Remarks
Greenburg-Smith impinger.	8	[1] 28.3	75–100	Favorable for sampling in low humidities. Commercially available.
All-glass impinger (Chemical Corps type).	9	[1] 6 and 12.5	10–20	Inlet tube affords some particle-size discrimination. Commercially available.
Pre-impinger	10	[1] 12.5	5	Separates particles into two size groups. Commercially available.
Capillary impinger	11	[1] 2–3	25	Adapted to high concentrations only. Not available commercially, readily constructed in laboratory, inexpensive.
Membrane filter impinger.	12	[1] 5	20	Adapted to low concentrations only. Not commercially available, expensive to construct.
Multiport impinger	13	[1] 12	20	Affords multidirectional sampling. Plastic construction. Not commercially available, expensive to construct.

Table 3. Liquid impingement samplers—Continued

Sampler	Figure number	Sampling rate (lpm)	Volume of collecting liquid (ml.)	Remarks
Tangential jet impinger:				
TDL	14	[1] 28.3	25	Ships eliminates particle retention in inlet tubes. TDL has higher sampling rate. Not commercially available, readily constructed in laboratory, inexpensive.
Ships		[1] 10	10	
Midget impinger	15	[2] 2.5–5	10	Adapted to high concentrations only, commercially available.
Folin bubbler	16	[2] 28.3	20	Components for construction are inexpensive and commercially available.
Venturi scrubber:				
Large	17	[1] 450–850	100	Large model favorable for sampling in low humidities. Not commercially available, expensive to construct.
Small		[1] 25	20	
Vigreaux bubbler	18	[2] 0.1–1	30	Adapted to high concentrations only. Not commercially available.
Syringe	19	[2] Approx. 5 ml./sec.	20	Adapted to high concentrations only. Suitable for sampling submicron particles. Commercially available.

[1] High velocity. [2] Low velocity.

Table 4. Filtration samplers

Common characteristics: Usefulness depends upon ability of micro-organisms to resist desiccation associated with filter collection. Suitable for collection of spores and other resistant microbial forms. High collection efficiency. Quantitation by washing and plating procedures except membrane filter. Some disintegration of particles can occur during washing. Final assay results expressed as organisms per unit volume of air. Can be used at temperatures above and below freezing. Simple to operate, portable, all filter media commercially available. All can be sterilized by gas, but only a few can be autoclaved.

Sampler	Figure number	Sampling rate (lpm)	Remarks
Membrane filter	20	5–28.3	Collects nondisintegrated or unmodified particles. No diluting or plating procedures required. Final assay results expressed as particles per unit volume of air. Can be made transparent for direct particle-size measurement. Not well adapted to high concentrations. Can be sterilized by autoclaving. Commercially available.
Cotton filter	21	10	Not well adapted to low concentrations. Can be sterilized by autoclaving. Components for construction are inexpensive and commercially available.
Respiratory	22	10 (sedentary)	Limited application. Not well adapted to low concentrations. Can be sterilized by autoclaving. Not commercially available, inexpensive to construct.
Modified high volume air	23	1,415 (50 c.f.m.)	Self-contained unit. High sampling rate. Large volume of liquid required for assay. Commercially available.
Modified Hemeon	24	6–10	Collects intermittent samples automatically. Renders a time-concentration relationship. Not well adapted to low concentrations. Commercially available.
In-line filter paper	25	10 (varies, depending on paper).	Not well adapted to low concentrations. Can be sterilized by autoclaving.
Gel-Foam	26	Variable	Special tools required for preparing filter pad. Nonuniform in structure. Cracking and channeling can occur. High resistance. Not commercially available.

Table 5. Impaction samplers

Common characteristics: Relatively high collection efficiency. Fabricated from metal; rugged, portable, simple to operate. Relatively high sampling volume. Some cannot be sterilized by autoclaving. Gaseous sterilization is desirable, although swabbing with disinfectant is often sufficient. Samplers employing agar limited to use in temperatures above 32° F. unless some method of heating is provided.

Sampler	Figure number	Sampling rate (lpm)	Collecting surface	Remarks
Cascade impactor	27	17.5	Glass slide, plain or coated.	Separates particles into four size ranges. Size distribution of particles can be determined from data. Limited to low concentration if particles are to be counted. Particle shattering may occur. Suitable for resistant organisms only, such as spores, if viable counts are to be made by bacteriological methods. Viable quantitation by washing and plating procedures. Final assay results expressed as organism per unit volume of air. Commercially available.
Single stage impactor (Johnson)	28	12.5	Glass disk or agar	Separates particles into two size groups. Particle size cutoff point can be varied by changing the diameter of the inlet orifice. Limited to low concentrations if particles are to be counted. Particle shattering may occur when glass disk is used. Suitable for vegetative forms when agar is used. Viable quantitation by washing and plating procedures. Final assay results expressed as organisms per unit volume of air. Not commercially available.
TDL cascaded slit (modified Casella, 4-stage).	29	28.3	Agar	Size distribution of particles can be determined from data. Collects nondisintegrated or unmodified particles. No dilution or plating procedures required. Not well adapted to high concentrations. Final assay results expressed as particles per unit volume of air. Single units commercially available, costly to assemble in cascade manner.
Fort Detrick slit	30	28.3	Agar	Renders time-concentration relationship. Collects nondisintegrated or unmodified particles. No diluting or plating procedures required. Not well adapted to high concentrations. Final assay results expressed as particles per unit volume of air. Adjustable slit permits varied flow rate Commercially available.

Table 5. Impaction samplers—Continued

Sampler	Figure number	Sampling rate (lpm)	Collecting surface	Remarks
Fort Detrick slit-incubator	31	14.2	Agar	Renders time-concentration relationship. Collects nondisintegrated or unmodified particles. No diluting or plating procedures required. Not well adapted to high concentrations. Final assay results expressed as particles per unit volume of air. Adjustable slit permits varied flow rate. Permits longer sampling periods than other slit samplers. Agar plate can be incubated without removing plate from sampler. Not commercially available, expensive to construct.
Slit (Casella)	32	28.3	Agar	Renders time-concentration relationship. Collects nondisintegrated or unmodified particles. No diluting or plating procedures required. Not well adapted to high concentrations. Final assay results expressed as particles per unit volume of air. Adjustable slit permits varied flow rate. Commercially available.
Sieve	33	28.3	Agar	Collects nondisintegrated or unmodified particles. No diluting or plating procedures required. Uses standard Petri dish. Limited to low concentrations. Superimposing of particles can occur. Limited to short sampling times. Final assay results expressed as particles per unit volume of air. Not commercially available, expensive to construct.
Andersen	34	28.3	Agar, glass, or metal.	Separates particles into six size ranges. Size distribution of particles can be determined from data. Collects nondisintegrated or unmodified particles. No diluting or plating procedures required. Not well adapted to high concentrations. Superimposing of particles can occur. Final assay results expressed as particles per unit volume of air. Commercially available.
Modified commercial respirator	35	10 (sedentary)	Agar	Collects nondisintegrated or unmodified particles. No diluting or plating procedures required. Limited to low concentrations. Not quantitative. Low collection efficiency. High resistance to breathing. Limited application. Respirator can be fabricated from commercial mask.

Table 6. Sedimentation samplers

Common characteristics: Long sampling time required for all particles to settle. Agglomeration of particles can occur during settling process. Generally selective for large particles. Results affected by air currents and drafts. Simple to use, inexpensive, readily available, easily sterilized by autoclaving. The particles are randomly distributed over the collecting surfaces. Collection of viable particles on non-nutrient surfaces is limited to spores and other resistant microbial forms.

Sampler	Figure number	Collecting surface	Remarks
Settling plates (agar)	36	Agar, membrane filter, glass plate, or slide.	Collects nondisintegrated or unmodified particles for direct microscopic examination or colony growth. Limited value for quantitative measurement of airborne particles. Viable quantitation by washing and plating procedures when glass surface is used. Final assay results expressed as organisms per unit area when glass surface is used.
Surface	37	Metal, cloth, or paper.	Collects nondisintegrated or unmodified particles. Viable quantitation by washing and plating procedures. Final assay results expressed as organisms per unit area.
Grab	38	Agar, membrane filter, or glass slide.	Collects nondisintegrated or unmodified particles for direct microscopic examination or colony growth. Provides a quantitative measure of the suspended particles. Only a small volume of air sampled. Viable quantitation by washing and plating procedures when glass surface is used. Final assay results expressed as organisms per unit volume of air when glass surface is used.

Table 7. Centrifugal samplers

Common characteristics: Collect nondisintegrated or unmodified particles for microscopic examination or viable quantitation. Particles are not randomly distributed over the collecting surfaces. Collection of viable particles on non-nutrient surfaces is limited to spores and other resistant microbial forms. Efficiency depends upon operating conditions and size of the particles. Can be sterilized by gas.

Sampler	Figure number	Sampling rate (lpm)	Collecting surface	Remarks
Wells air centrifuge	39	28–42	Agar or liquid	Motor, flowmeter, and sampler are combined in one unit. When agar is used, sampling is limited to low concentrations, no diluting and plating procedures are required, colony counting is difficult and colonial morphology is not easily studied. Final assay results expressed as particles per unit volume of air. Not commercially available.
Aerotec tube	40	28.3	Glass slide or vial.	Separates particles into two size groups. Particle-size cutoff point can be varied by changing the airflow. Limited to low concentrations if particles are to be counted. Viable quantitation by washing and plating procedures. Final assay results expressed as organisms per unit volume of air. Commercially available.
Conifuge	41	0.025	Glass slide or membrane filter.	Particles are distributed over collecting surface according to their mass or size. Size distribution of particles can be determined from data. Low sampling rate. Viable quantitation by washing and plating procedures when glass surface is used. Final assay results expressed as organisms per unit volume of air when glass surface is used. Commercially available.

Table 8. Electrostatic and thermal precipitation samplers

Sampler	Figure number	Sampling rate (lpm)	Collecting surface	Remarks
Electrostatic precipitator	42	28.3	Agar	Self-contained unit. Collects nondisintegrated or unmodified particles for colony growth. No diluting or plating procedures required. Not well adapted to high concentrations. Final assay results expressed as particles per unit volume of air. High sampling rate and collection efficiency. Low resistance to airflow. Complex, requires careful handling. Can be sterilized by gas. Commercially available.
Thermal precipitator: Water cooled	43	0.4	Glass	Self-contained unit. Collects nondisintegrated particles for microscopic counting and sizing. Size distribution of particles can be determined from data. Limited to low concentrations if particles are to be counted. Not generally used for quantitation of viable particles. High collection efficiency for small particles (less than 5 microns). Low sampling rate. Complex, requires careful handling and adjustment. Can be sterilized by gas. Commercially available.
Hot wire	44	0.007	Glass coverslip	Collects nondisintegrated particles for microscopic counting and sizing. Size distribution of particles can be determined from data. Limited to low concentrations if particles are to be counted. Not generally used for quantitation of viable particles. High collection efficiency for small particles (less than 5 microns). Very low sampling rate. Complex, requires careful handling and adjustment. Can be sterilized by gas. Commercially available.

Liquid Impingement Samplers

GREENBURG-SMITH IMPINGER

General Description: The Greenburg-Smith sampler is a high velocity impinger which has been used primarily in air pollution studies. The inlet of the standard instrument is located at the top with the vacuum connection immediately below. The tip of the inlet tube is drawn into an orifice and a glass plate is fixed at a distance of 5 mm. from the orifice. Incoming particles are thus impinged onto the plate and are momentarily arrested, after which they are washed into the liquid. The amount of liquid in the flask can vary, provided the plate is submerged a minimum of 3 cm. below the liquid level.

Optimum Flow Rate: 28.3 liters per minute.

Collection Surface: Organisms impinged on glass and washed into liquid collecting medium (75 to 100 ml.).

Source: Wilson Products, Inc., Industrial Hygiene Department, Reading, Pa. Drawings also available from Technical Development Laboratories, Communicable Disease Center, U.S. Public Health Service, P.O. Box 769, Savannah, Ga.

References: 7 and 8.

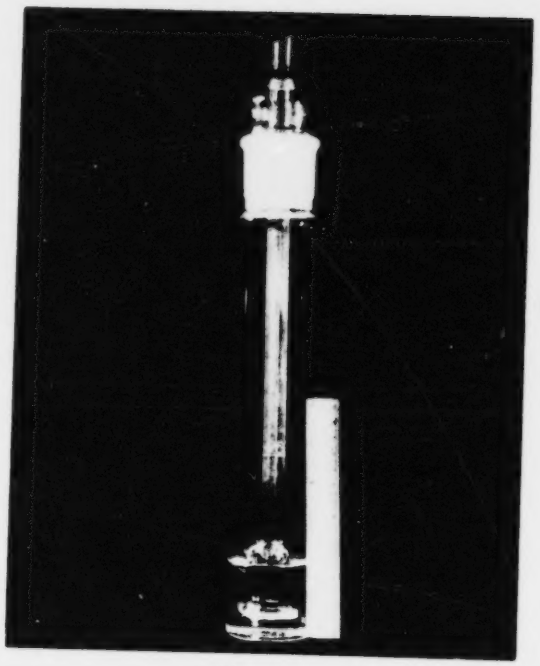


Figure 8

ALL-GLASS IMPINGER (Chemical Corps Type)

General Description: This high velocity impinger is a modification of the Greenburg-Smith impinger. The upper end of the inlet tube is bent into a 90° curve of large radius, and the stem and flask are fitted together through a ground glass connection. A short length of capillary tubing is fused to the lower end of the inlet tube so that the tip is 4 mm. from the flask bottom, while in another model, the stem is 30 mm. from the flask bottom. The 30 mm. raised stem provides an increase in the collection of viable vegetative cells. The vacuum outlet arrangement is similar to that of the Greenburg-Smith sampler. This sampler can be operated with a pre-impinger discussed on page 25.

Optimum Flow Rate: 6 and 12.5 liters per minute.

Collection Surface: Liquid (20 ml.).

Source: Ace Glass, Inc., Vineland, N.J.

Reference: 6.

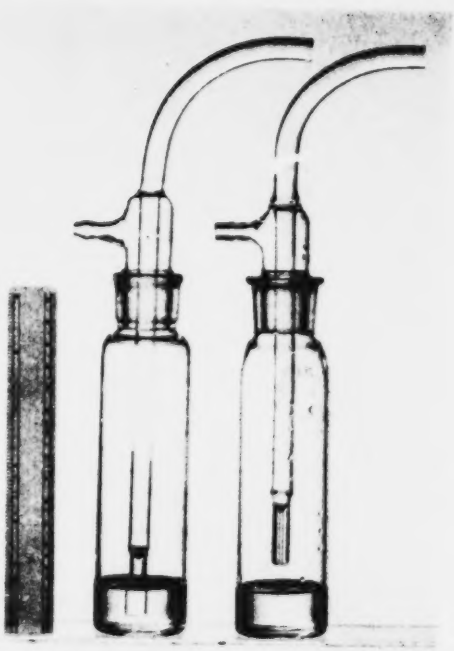


Figure 9.

PRE-IMPINGER

General Description: The pre-impinger is a spherical device which may be fitted to the inlet tube of a liquid impinger or equivalent sampler operated at the same flow. This sampler divides the total aerosol into two particle-size fractions, the cutoff being above and below 5 microns. The large particles (> 5 μ) are retained within the pre-impinger and the small particles pass through and are collected in the impinger. Bacteriological counts of the number of organisms entrapped in the pre-impinger (> 5 μ) are determined by standard diluting and plating procedures. If the organisms collected in the pre-impinger are not to be enumerated, the pre-impinger can be operated without liquid. In such instances the particles greater than 5 microns will be impinged on the interior surface of the pre-impinger.

Optimum Flow Rate: 12.5 liters per minute.

Collection Surface: Liquid (5 ml.) or on the interior glass surface.

Source: Chatas Glass Co., Vineland, N.J.

Reference: 66.

Figure 10.

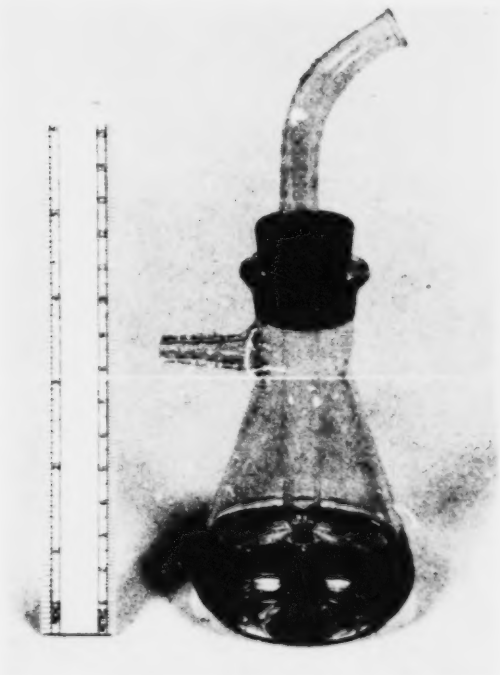


Figure 11.

CAPILLARY IMPINGER

General Description: This high velocity impinger is useful in aerosol chamber work where sampling flow rates may be only 2 or 3 liters per minute. There are several models of the capillary impinger currently in use. The impinger described consists of a short length of 0.558-mm. (inside diameter) capillary tubing fused to an inlet tube which has a 90° bend at the top. The tube is inserted through a rubber stopper and adjusted so that when the assembly is placed in a 125-ml. filtering flask the tip of the capillary is 4 mm. from the bottom of the flask. When 25 ml. of sampling fluid is added to the flask, the fluid level is approximately 5 mm. above the capillary tip. The sampler is then operated so that the capillary tip acts as a critical orifice.

Optimum Flow Rate: 2 to 3 liters per minute.

Collection Surface: Liquid (25 ml.).

Source: No commercial source. Drawings available from Technical Development Laboratories, Communicable Disease Center, U.S. Public Health Service, P.O. Box 769, Savannah, Ga.

Reference: 77.

MEMBRANE FILTER IMPINGER

General Description: The liquid membrane filter sampler combines the operating principles of the liquid impinger and membrane filter samplers. Organisms are trapped in the collecting fluid by impingement when air is drawn through the inlet orifice. After sampling, the collecting fluid is filtered through the membrane filter located in the top of the sampler by inverting the sampler and applying a vacuum to the outlet tube. The filter is removed and cultured by standard membrane filter techniques. Quantitative results cannot be obtained by this impinger when sampling high concentration aerosols since overcrowding of the membrane filters will occur. Wetting of the filter due to excessive bubbling of the collecting fluid increases airflow resistance during operation. Airflows up to 5 liters per minute are suitable with 20 ml. of collecting fluid.

Optimum Flow Rate: 5 liters per minute.

Figure 12.

Collection Surface: Liquid (20 ml.) and filter.

Source: Sampler—No commercial source. Drawings available from Technical Development Laboratories, Communicable Disease Center, U.S. Public Health Service, P.O. Box 769, Savannah, Ga.
Membrane filter, Type HA: Millipore Filter Corp., Watertown 72, Mass.
Type Bac-T-Flex: Carl Schleicher & Schuell Co., Keene, N.H.

MULTIPORT IMPINGER

General Description: This sampler consists of a 5.08-cm. (2 inch) diameter plastic barrel with four orifices drilled tangentially 1 inch above the bottom and sloping downward at a 45° angle. The orifices are spaced 90° apart around the sides of the barrel. Air enters the orifices and impinges in the liquid, then leaves the sampler through a cone type baffle at the top. The cone is made of plastic and is sealed with a rubber O ring gasket. After sampling, the liquid is assayed by standard bacteriological techniques.

Optimum Flow Rate: 12 liters per minute.

Collection Surface: Liquid (20 ml.).

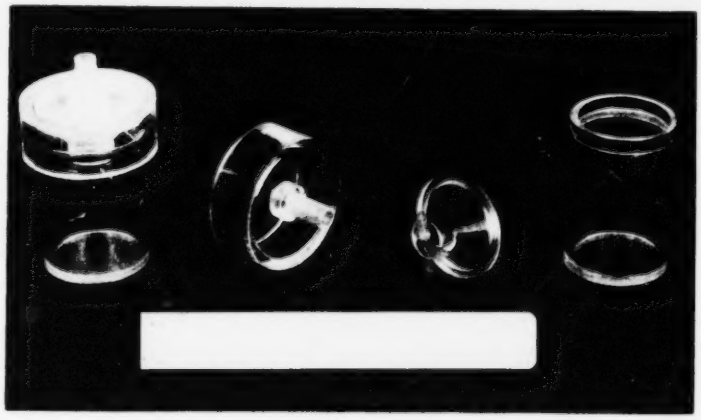


Figure 13.

Source: No commercial source. Drawings available from Technical Development Laboratories, Communicable Disease Center, U.S. Public Health Service, P.O. Box 769, Savannah, Ga.

TANGENTIAL JET IMPINGER (TDL and Ships)

General Description: This sampler is a high velocity type liquid impinger in which sampled air is drawn through an aluminum or stainless steel jet or orifice located near the base tangentially to the circumference of the flask and inclined downward at a 20° angle with the base. The outlet port is located in the rubber or ground glass stoppered top. The sampler is operated so that the inlet jet operates as a critical orifice.

Optimum Flow Rate: 28.3 liters per minute (TDL model); 70 liters per minute (Shipe).

Collection Surface: Liquid 25 ml. (TDL model); 10 ml. (Shipe model).

Source: No commercial source. Drawings available from Technical Development Laboratories, Communicable Disease Center, U.S. Public Health Service, P.O. Box 769, Savannah, Ga.

Reference: 10.

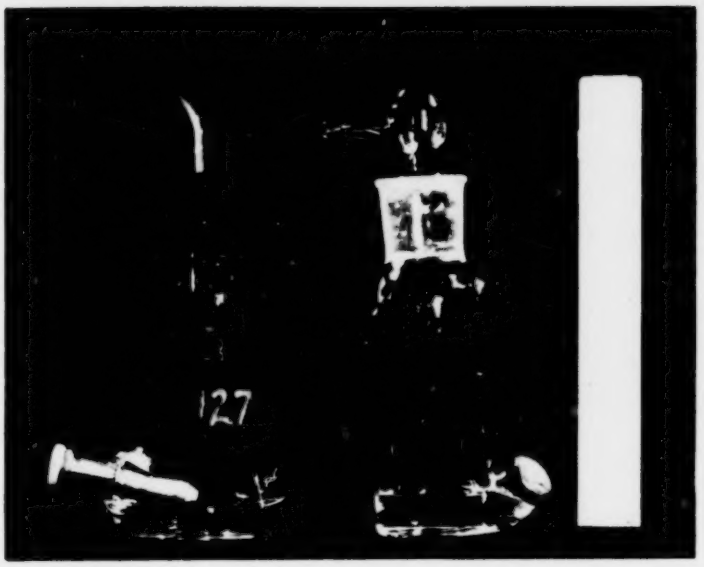


Figure 14.

MIDGET IMPINGER

General Description: This impinger is used for sampling small air volumes at a low jet velocity. A straight inlet tube, inserted through a rubber stopper, fits in a 15.2-cm. × 2.5-cm. graduated cylinder. The lower end of the inlet tube terminates in a 1-mm. diameter orifice adjusted to a point 5 mm. above the bottom of the cylinder.

Optimum Flow Rate: 2.5 to 5 liters per minute.

Collection Surface: Liquid (10 ml.).

Source: Mine Safety Appliance Co., 201 North Braddock Avenue, Pittsburgh 8, Pa., or from any scientific laboratory supply house.

References: 78 and 79.

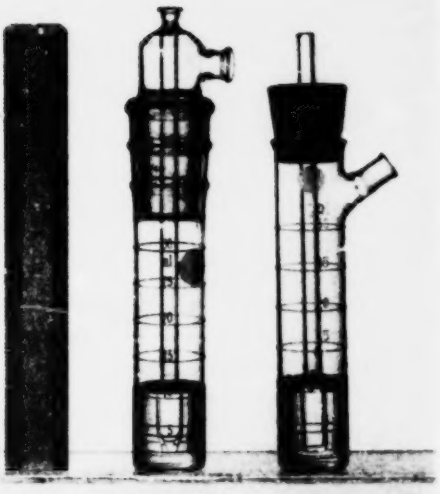


Figure 15.

FOLIN BUBBLER

General Description: The low velocity Folin bubbler is the same as the Folin aeration tube which is available at any scientific supply house. Six small holes are bored horizontally into and around a glass inlet bulb located at one end of the tube. The tube is inserted in a rubber stopper and this assembly is placed in any type flask desired. Test tubes 20 cm. long and 2.5 cm. in diameter have been used as flasks. An antifoam agent and a Kjeldahl trap are recommended to minimize liquid loss.

Optimum Flow Rate: 28.3 liters per minute.

Collection Surface: Liquid (20 ml.).

Source: Components are commercially available from any scientific laboratory glassware supplier. Construction described in reference.

Reference: 11.

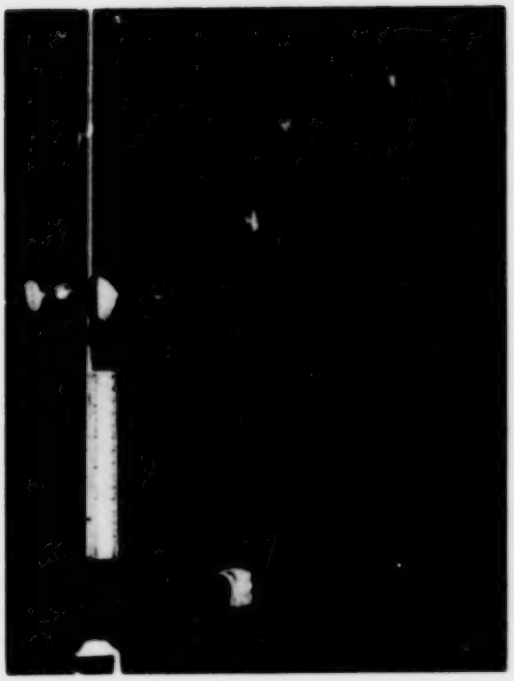


Figure 16.

VENTURI SCRUBBER

General Description: The venturi scrubber is a laboratory adaptation of a large industrial type air cleaning device. There are two models available. An airflow of 470 liters per minute at a vacuum of 6.7 cm. of mercury is attained with the large model when operated with a household vacuum cleaner blower. Maximum attainable flow (critical) is between 715 and 850 liters per minute. Incoming air passes around the nozzle of the collecting fluid line and through the throat of the venturi where a reduced pressure causes the fluid to flow into the venturi. As the collecting fluid leaves the nozzle, it is broken into a spray by the shearing action of the airstream. The liquid with entrapped particles settles into the bowl and promptly recirculates. The recycling of the fluid breaks up particles so that single cells are suspended in the collection bowl.

Optimum Flow Rate: Large model, 450 to 850 liters per minute; small model, 25 liters per minute.

Collection Surface: Liquid (large model, 100 ml.; small model, 20 ml.).

Source: No commercial source. Drawings available from Technical Development Laboratories, Communicable Disease Center, U.S. Public Health Service, P.O. Box 769, Savannah, Ga.

Reference: 12.

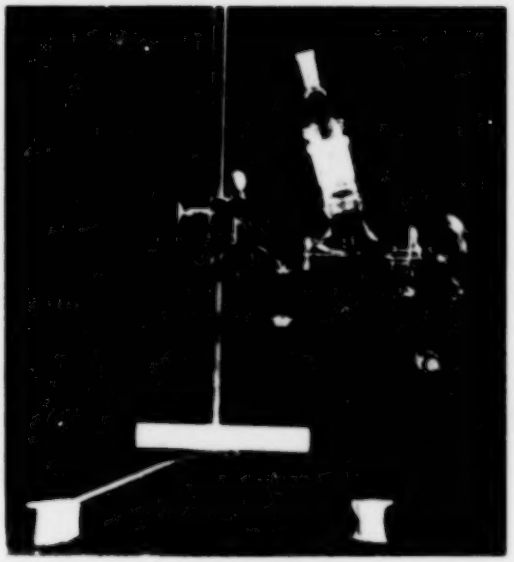


Figure 17.

VIGREAUX BUBBLER

General Description: The Vigreaux bubbler was modified for the collection of airborne *Escherichia coli* T-3 phage aerosols (0.045 microns) and consists of a distilling column modified for air sampling. Indentations in the wall of the column increase the contact area and time between the sampled air and the collecting fluid. Phage particles are relatively unstable in an aerosol state, especially at low relative humidities. The sampling efficiency of the Vigreaux bubbler is a function of the airflow rate. A flow rate of 1.0 liter per minute is most efficient for the collection of phage aerosols at relative humidities of 70 percent and higher. At 30 percent relative humidity, a flow rate of 0.15 liters per minute or less must be used to maintain a high efficiency. Although this sampler is designed primarily for the collection of phage aerosols, it can be used for the collection of airborne bacteria.

Optimum Flow Rate: 0.1 to 1 liter per minute.

Collection Surface: Liquid (30 ml.).

Source: No commercial source. Drawings available from Technical Development Laboratories, Communicable Disease Center, U.S. Public Health Service, P.O. Box 769, Savannah, Ga.

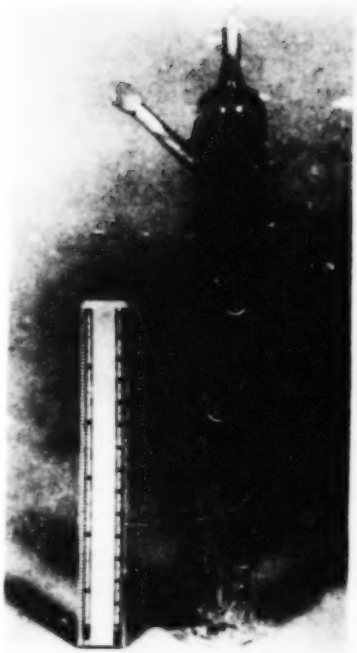


Figure 18.

SYRINGE SAMPLER

General Description: The syringe sampler consists of a 100-ml. hypodermic syringe containing 20 ml. of collecting liquid and is operated by bubbling 50 ml. of aerosol through the collecting fluid in 10 seconds. The sample is shaken for at least 1 minute and subsequently assayed according to standard bacteriological methods.

Optimum Flow Rate: 5 ml. per second.

Collection Surface: Liquid (20 ml.).

Source: Becton, Dickinson & Co., Rutherford, N.J.

Reference: 80.

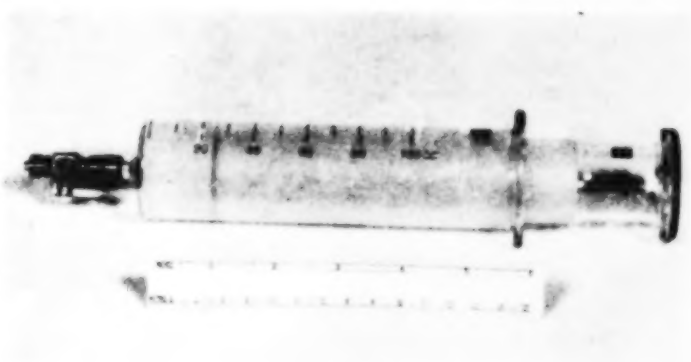


Figure 19.

MEMBRANE FILTER

General Description: Molecular membrane filters (fig. 20) developed for aerosol assay consist of stabilized dry, cellulose ester-gel membranes approximately 150 microns thick. Several standard sizes are commercially available, and range from 47 mm. to 300 mm. in diameter. Other sizes are readily available on special order. Two types of filters are available, one for microbiological study of gases (Type AA and Coarse, Group 2), the other for microbiological study of liquids (Type HA and Bac-T-Flex). The average pore size of the AA Millipore filter is 0.80 $\mu \pm 0.05$ μ. This permits an airflow of 33,000 cc./sq. cm./min. at 25 C. and 70 cm. Hg. The HA Millipore filter (0.45 $\mu \pm 0.02$ μ pore size) permits an airflow of 9,600 cc./sq. cm./min. at 25 C. and 70 cm. Hg. The pore size and resistance of other commercial membrane filters are similar to that specified above. The membrane is supported on a porous carbon disk or fine stainless steel screen in a holder, such as is illustrated (fig. 45, p. 46). After the air sample has been collected, the filter is placed in a plastic dish containing a blotter pad previously wetted with a small amount of a nutrient broth. The membrane filter may also be cultured on an agar surface. Following incubation, the resulting colonies are counted with the aid of a stereoscopic microscope.

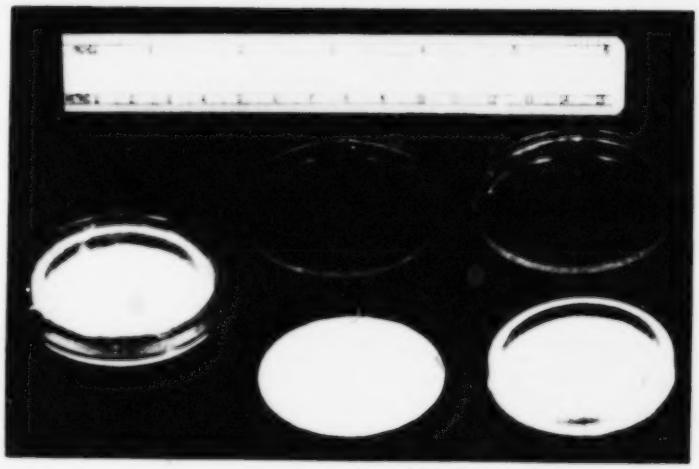


Figure 20.

Optimum Flow Rates: Normally used at flows of 5 to 28.3 liters per minute.

Collection Surface: Membrane filter.

Sources: Filters: Unringed, HA and AA; Ringed, HA and AA—Millipore Filter Corp., Watertown 72, Mass.
Unringed, Bac-T-Flex; Coarse, Group 2—Carl Schleicher & Schuell Co., Keene, N.H.

References: 23, 24, and 25.

COTTON FILTER

General Description: The cotton sampler consists of a modified glass drying tube, a glass inlet tube, rubber stopper, wire screen, and absorbent cotton. The drying tube is filled approximately two-thirds full of absorbent cotton which is packed to a resistance of 38 cm. of water at a flow rate of 10 liters per minute. The wire screen placed in the bottom of the tube prevents the cotton from being pulled through the sampler. The sampler is operated by applying the vacuum source to the outlet tube. After collection of the aerosol, the cotton is removed, placed into a bottle containing 50 or 100 ml. of sterile distilled water, shaken for 10 minutes, and biologically assayed. In lieu of cotton, alginate wool may be used.

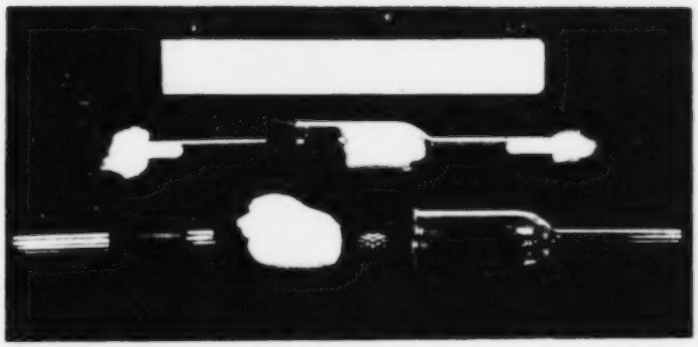


Figure 21.

Optimum Flow Rate: 10 liters per minute.

Collection Surface: Absorbent cotton.

Source: No commercial source. Construction shown. reference. Glass drying tube—ACE Glass Co., Inc., Vineland, N.J.

Reference: 38.

RESPIRATORY SAMPLER

General Description: This sampler, designed to sample inspired bacteria, is composed of a rubber mouthpiece into which is inserted a cartridge containing cotton packed to a resistance of 25 mm. of water at a flow of 10 liters per minute. The subject inhales through the sampler and exhales through his nose. After use, the cotton plug is removed and shaken in 25 ml. of sterile water or other suitable liquid to resuspend the collected organisms. Aliquots are taken and plated according to standard techniques for biological assay.

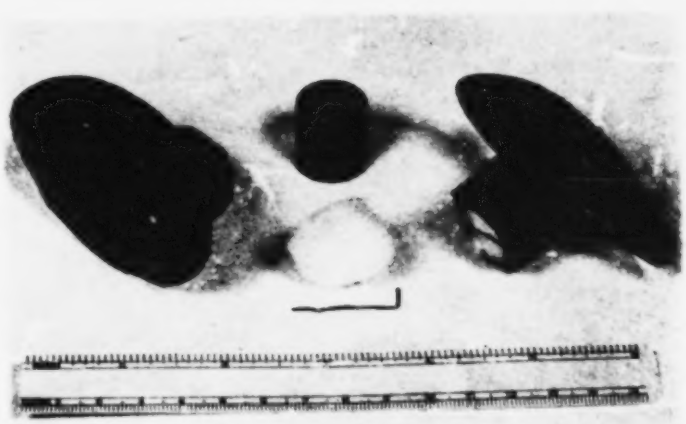


Figure 22.

Optimum Flow Rate: Breathing rate of users, approximately 10 liters per minute at sedentary conditions.

Collection Surface: Absorbent cotton.

Source: Mouthpiece — Sanborn Corp., 175 Wyman Street, Waltham, Mass. Drawings available from Technical Development Laboratories, Communicable Disease Center, U.S. Public Health Service, P.O. Box 769, Savannah, Ga.

Reference: 38.

MODIFIED HIGH VOLUME AIR SAMPLER

General Description: Air is drawn through a 20.3-cm. × 25.4-cm. (8 inch × 10 inch) sheet of filter paper by a blower connected to the paper holder. After the sample is collected the paper is removed and shaken in 250 ml. of sterile water or other suitable fluid to resuspend the organisms which are then assayed according to standard bacteriological methods.

Optimum Flow Rates: Approximately 50 cubic feet per minute.

Collection Surface: Filter paper.

Sources: Sampler—Atomic Center, Lake Avenue, Center Moriches, Long Island, N.Y., and Mine Safety Appliance Co., 201 North Braddock Avenue, Pittsburgh 8, Pa.

Figure 23.

Filter paper (Glass Fibre Filter Web, Catalogue Nos. CT75428 and CT26066): Mine Safety Appliance Co., 201 North Braddock Avenue, Pittsburgh 8, Pa.

References: 31, 37.

MODIFIED HEMEON SAMPLER

General Description: This sampler (frequently called the American Iron and Steel Institute sampler) uses a diaphragm type pump to draw air through filter paper in tape form and provides a means of taking 30-minute samples automatically over an 8-hour period. The tape is clamped between two parts of the inlet tube so that all air must pass through the filter. At predetermined intervals the inlet tube separates, the filter tape moves forward to a new sampling position, and sampling is resumed. The sampler has been modified to allow the filter samples to lie flat in a cassette to prevent disturbance of the collected particles. When sampling is completed, each section of the filter is cut off, placed in bottles of sterile liquid, and shaken to suspend the organisms which are then assayed according to standard bacteriological methods.

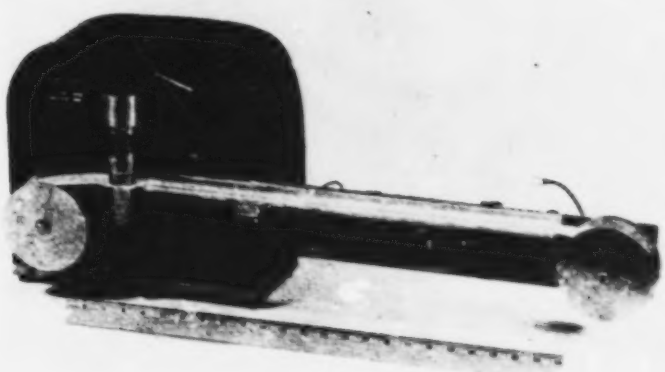


Figure 24.

Optimum Flow Rates: 6–10 liters per minute. Exact flow depends on filter medium selected.

Collection Surface: Filter paper.

Sources: Sampler — Research Appliance Co., P.O. Box 307, Allison Park, Pa.
Filter paper (Glass Fibre Filter Web Catalogue No. CT75428): Mine Safety Appliance Co., 201 North Braddock Avenue, Pittsburgh 8, Pa.

References: 31, 32, and 33.

IN-LINE FILTER PAPER SAMPLER

General Description: This sampler is composed of two 31.7-mm. diameter aluminum cones which screw together sealing a 25.4-mm. (1 inch) diameter disk of filter paper between the hollow cones. One-fourth-inch diameter tubes at each end of the cones are used for vacuum connection and air inlets. After use, the paper is removed, shaken in sterile water, and then assayed according to standard bacteriological methods.

Optimum Flow Rate: 10 liters per minute. Exact flow rate depends on filter paper selected.

Collection Surface: Filter paper.

Source: No commercial source. Drawings of this sampler are available from Technical Development Laboratories, Communicable Disease Center, U.S. Public Health Service, P.O. Box 769, Savannah, Ga.
Filter paper (Glass Fibre Filter Web Catalogue No. CT75128): Mine Safety Appliance Co., 201 North Braddock Avenue, Pittsburgh 8, Pa.

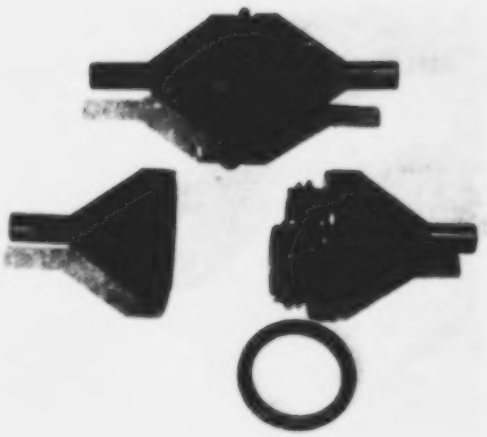


Figure 25.

GEL-FOAM SAMPLER

General Description: The Gel-Foam sampler consists of two funnel-shaped holders between which a Gel-Foam pad and its supporting fine mesh screen are placed. A cam and lever operated compression device seals the filter pad between the holders insuring the filtration of all sampled air. An orifice is located in the inlet tube between two pressure taps which serves as a flow metering device when connected to a manometer. A vacuum source, attached to the bottom, draws the sampled air through the filter.

When sampling is completed, the pads are dissolved by immersing in sterile water, agitating, and heating slightly (100° F.). The resulting suspension is then plated on a suitable agar.

Optimum Flow Rates: Commercial Gel-Foam varies considerably in airflow resistance, therefore specific flow at a given resistance cannot be quoted. Resistance normally is a minimum of several inches of mercury at airflows of 1 to 2 liters per minute.

Collection Surface: Water soluble Gel-Foam.

Source: Sampler and cutting tools are custom made. Filter material: Gel-Foam bricks (nonsterile), Upjohn Co., Kalamazoo, Mich.

References: 39 and 40.

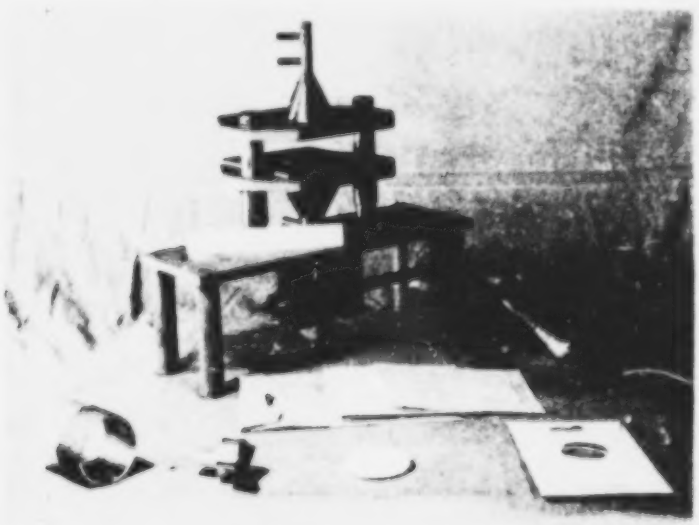


Figure 26.

CASCADE IMPACTOR (Casella

General Description: Air drawn into the cascade impactor is passed through four slit-type jets arranged in series. Each jet is directed against a glass slide. The successive jets decrease in area so that at each stage the sampled air is accelerated to a higher velocity. This results in particles of decreasing size range being impacted on successive slides.

The clearance between the face of the jet and the slide also varies. The first jet is chamfered so that there is a clearance of 1 mm. on the upstream side and 6 mm. on the downstream side. The second jet is chamfered at 45° with the lower edge nearly touching the slide. The third and fourth jets are not chamfered, and have clearances approximately equal to the slot width. A filter paper sampler may be added to serve as a fifth stage.

The inlet tube terminates in the first jet. The dimensions of the jet, approximate speed of the air passing through the jet, mass median diameter (MMD), and maximum particle size impacted on the slides by each jet at an airflow rate of 17.5 liters per minute are shown in the table.

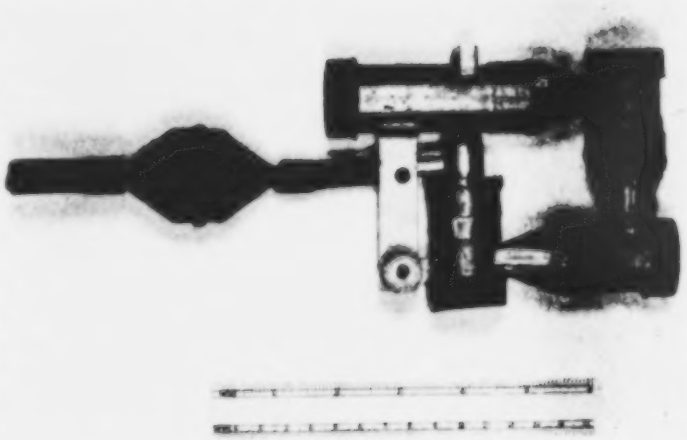


Figure 27.

Optimum Flow Rate: 17.5 liters per minute.

Collection Surface: Glass microscope slides.

Source: Catalogue No. CT34232: Mine Safety Appliance Co., 201 North Braddock Avenue, Pittsburgh 8, Pa.

References: 13 and 14.

Jet	*Dimensions (cm.)*	*Air speed meters/sec.*	*Mass median diameter[1] (microns)*	*Maximum particle size of each jet (microns)*
1	1.9 × (0.6–0.7)	2.4	—	200
2	1.4 × 0.20	11.1	12	20
3	1.4 × 0.075	28.8	4	7
4	1.4 × 0.027	83.0	2	2.5

[1] MMD for No. 1 jet depends on maximum size present in aerosol

SINGLE-STAGE IMPACTOR (Johnson)

General Description: This sampler, designed to separate aerosol particles into two size ranges, consists of a 6.5-cm. × 6-cm. × 3½-cm. aluminum box fitted with a threaded aluminum plug through which is drilled an orifice of proper diameter. The diameter of the orifice determines the size ranges into which the particles are separated. The plug has vertical extensions or legs to hold a glass slide or small dish of agar at the correct distance below the orifice for impaction of large particles. Small particles pass through the unit and are collected in an all-glass liquid impinger. The agar medium or glass microscope slide and the impinger liquid are assayed by standard techniques.

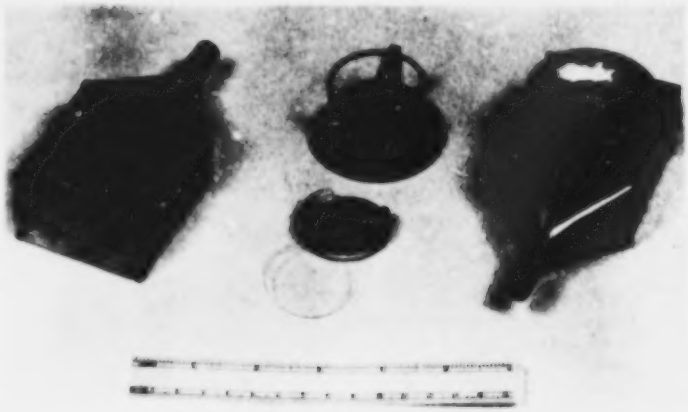


Figure 28.

Optimum Flow Rate: 12.5 liters per minute.

Collection Surface: Glass slide, or agar.

Source: No commercial source. Drawings available from Technical Development Laboratories, Communicable Disease Center, U.S. Public Health Service, P.O. Box 769, Savannah, Ga.

TDL CASCADED SLIT (Modified Casella, 4-Stage)

General Description: This sampler consists of four modified Casella (TDL) slit samplers arranged in series. Air passing through each slit is directed against the surface of agar in a rotating Petri dish. Each successive slit is decreased in area so that at each stage, the sampled air is accelerated to a higher velocity. This results in particles of decreasing size range being impacted on successive agar surfaces. After sampling, the Petri dishes are incubated and the colonies are counted. The colony counts on the successive stages provide information for viable particle-size distribution determinations. Each stage of this instrument can be removed and used separately as an individual slit sampler.

Optimum Flow Rate: 28.3 liters per minute.

Collection Surface: Agar.

Source: No commercial source. Sampler is in experimental stage. Drawings available from Technical Development Laboratories, Communicable Disease Center, U.S. Public Health Service, P.O. Box 769, Savannah, Ga.

Reference: Publication pending.

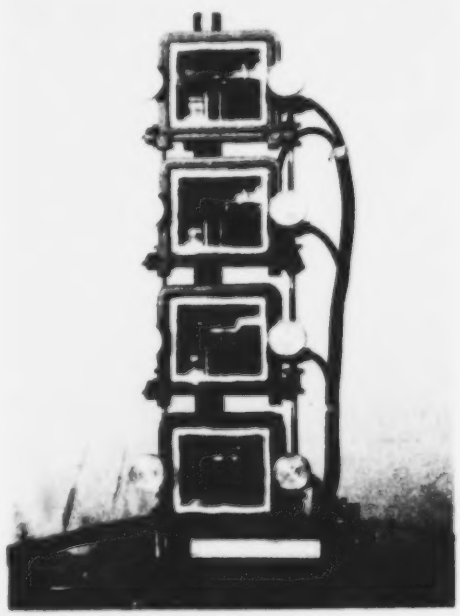


Figure 29.

FORT DETRICK SLIT SAMPLER

General Description: The sampler is a cylindrical unit having as its chief components a slit tube, agar culture plate, and interval timer. The slit tube is threaded into the top of the sampler and may be easily adjusted to the proper level (2–3 mm. above agar surface) as determined by a height indicator. The slit width is set at 0.152 mm. by an adjustable plate on the bottom of the slit tube. A 150-mm. × 20-mm. commercially available culture plate containing agar is rotated by means of a clock mechanism. The rate of rotation depends on selection of a commercial timer (30 minutes to 12 hours). Air is drawn through the slit, impacting the suspended particles on the rotating agar surface. After sampling, the plate is incubated and the colonies counted.

Optimum Flow Rates: 28.3 liters per minute. Flow rate may be varied by adjustment of slit width and vacuum supply.

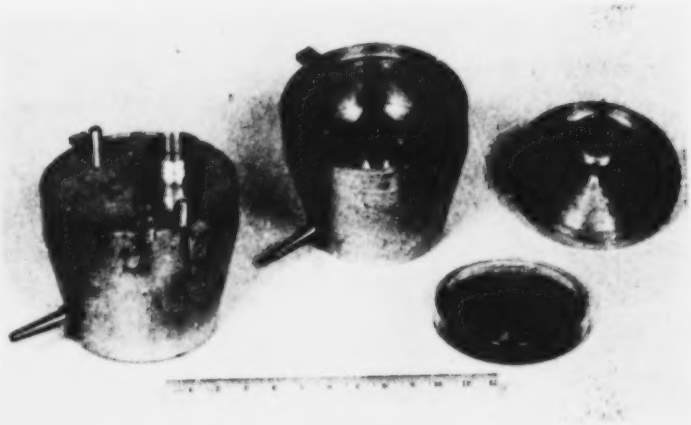


Figure 30.

Collection Surface: Agar.

Sources: Reynier and Sons, 3806 North Ashland Avenue, Chicago 13, Ill. Sampler drawings available from Technical Development Laboratories, Communicable Disease Center, U.S. Public Health Service, P.O. Box 769, Savannah, Ga.

Culture plates: Glass, any laboratory supply house; Plastic, Falcon Plastic Products, Culver City, Calif.

Reference: 19.

FORT DETRICK SLIT-INCUBATOR SAMPLER

General Description: The sampler is contained in a rectangular stainless steel box with a plastic (Plexiglas or Lucite) cover in the center of which is threaded an adjustable slit tube. The slit tube is adjusted to the proper level (2–3 mm. above the agar surface) as determined by the height indicator. The slit width is set at 0.102 mm. by an adjustable plate on the bottom of the slit tube. Air is drawn through the slit and the suspended particles are impacted on the surface of the agar contained in a moving rectangular metal tray (45.6 cm. × 6.35 cm.). The tray containing the medium is propelled under the slit at a rate of 5.71 cm. per hour by a rotating screw, powered by an electric clock motor. After the sample is collected, the plate enters the incubator section and the incubation cycle is started. Temperature is maintained by an electrically heated tape and is regulated with a thermostat. After incubation, the developed colonies are counted. This sampler renders a time concentration relationship and operates over a continuous 8-hour sampling period.

Optimum Flow Rate: 14.2 liters per minute.

Collection Surface: Agar.

Source: No commercial source. Drawings available from Technical Development Laboratories, Communicable Disease Center, U.S. Public Health Service, P.O. Box 769, Savannah, Ga.

Reference: 20.

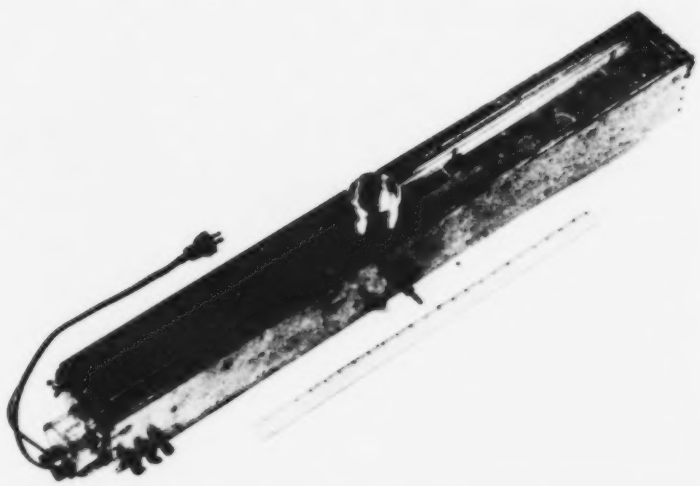


Figure 31.

SLIT SAMPLER (Casella)

General Description: The Casella slit sampler consists of an airtight chamber enclosing a rotating platform upon which is placed a Petri plate containing an appropriate medium. Air enters through a slit (0.33 mm. wide) located at the top of the chamber. Particles in the airstream are impacted on the agar surface located directly below the slit opening. The air leaves the chamber through an opening in the base or wall. The distance between the slit and agar surface is controlled by raising or lowering the rotating platform with an arm and screw adjustment. The rotating platform can be operated at three different rotation speeds ($\frac{1}{2}$, 2, and 5 rpm) by an electrically driven mechanism located directly below the chamber. The Casella sampler has a small opening, at the bottom of the airtight chamber, which is connected to a manometer for measuring airflow through the slit.

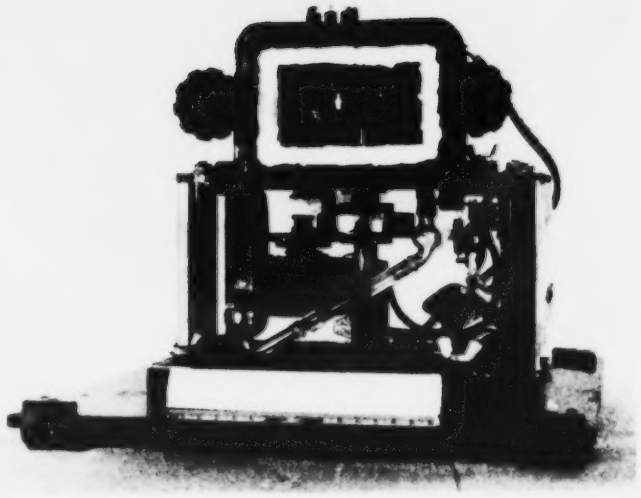


Figure 32.

Optimum Flow Rate: 28.3 liters per minute.

Collection Surface: Agar.

Sources: C. F. Casella and Co., Ltd., Regent House, Fitzroy Square, London, England.
Distributed by: C. A. Brinkmann and Co., 370 Great Neck Road, Great Neck, Long Island, N.Y.

Similar sampler available from Elliott Engineering Co., 649 Ponce de Leon Avenue, Decatur, Ga.

References: 16 and 17.

SIEVE SAMPLER

General Description: The sieve sampler consists of an aluminum container that holds a standard Petri dish. The container is fitted with a cover, the inner edge of which is threaded to receive a sieve plate with 340 holes (No. 79 drill) drilled equal distances from each other over its surface. The center of the sieve plate contains a gauge pin which is used to indicate the distance (2 to 4 mm.) between the sieve plate and the agar surface. Air is drawn through the small holes and particles are impacted on the agar surface. The sieve sampler can be equipped with an inverted funnel so that samples may be taken through small openings from an aerosol chamber or from air ducts, etc. However, use of the funnel may decrease the sampler efficiency. The sampler may be operated in any position.

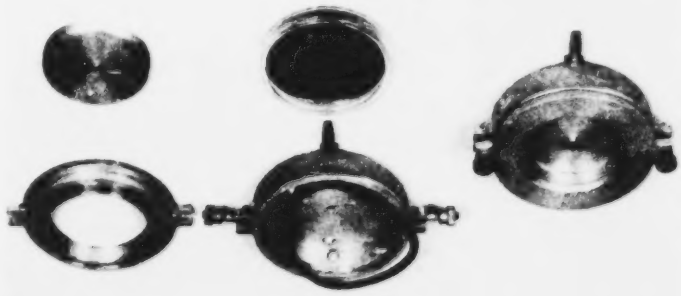


Figure 33.

After sampling, the plate is incubated and the colonies are counted.

Optimum Flow Rate: 28.3 liters per minute.

Collection Surface: Agar.

Source: No commercial source. Construction described in reference 15. Drawings also available from Technical Development Laboratories, Communicable Disease Center, U.S. Public Health Service, P.O. Box 769, Savannah, Ga.

References: 15 and 16.

ANDERSEN SAMPLER

General Description: The Andersen sampler is essentially a cascaded arrangement of sieve type samplers which have smaller diameter holes in each succeeding plate. The six-stage unit has hole diameters, by stage, as follows: 1 1.81 mm. (0.046 inch), 2 0.91 mm. (0.036 inch), 3 0.71 mm. (0.028 inch), 4 0.53 mm. (0.021 inch), 5 0.343 mm. (0.013 inch), and 6 0.25 mm. (0.010 inch). The distance of the collecting surface from the holes is controlled by utilizing a specially designed Petri dish and a specific quantity of agar media (27 ml.). Air is drawn successively through the six stages at increasing velocity so that the larger particles are impacted on agar surfaces in the first stages, the smaller particles, depending on their size and inertia are impacted on the last stages. After the sample has been taken, the plates are removed and incubated. For non-viable aerosol studies, the particles can be collected on glass or stainless steel surfaces.

Figure 34.

Optimum Flow Rate: 28.3 liters per minute.

Collection Surface: Agar, glass, or metal.

Source: A. A. Andersen, 1074 Ash Avenue, Provo, Utah.

References: 21.

MODIFIED COMMERCIAL RESPIRATOR SAMPLER

General Description: This commercial dust respirator is modified to permit sampling of air by the sieve sampler technique. The air enters the mask through a plastic disk containing 600 evenly spaced holes (0.368-mm. diameter) and the particles are impacted on an agar medium (10 ml.) contained in a small plastic Petri dish (57.1 mm. in diameter and 12.7 mm. in depth with cover).

Optimum Flow Rate: Breathing rate of user (approximately 10 liters per minute sedentary).

Collection Surface: Agar.

Source: No commercial source. Sampler may be fabricated from drawings and commercially available mask. Drawings available from Technical Development Laboratories, Communicable Disease Center, U.S. Public Health Service, P.O. Box 769, Savannah, Ga.
Mask: American Optical Co., Southbridge, Mass.

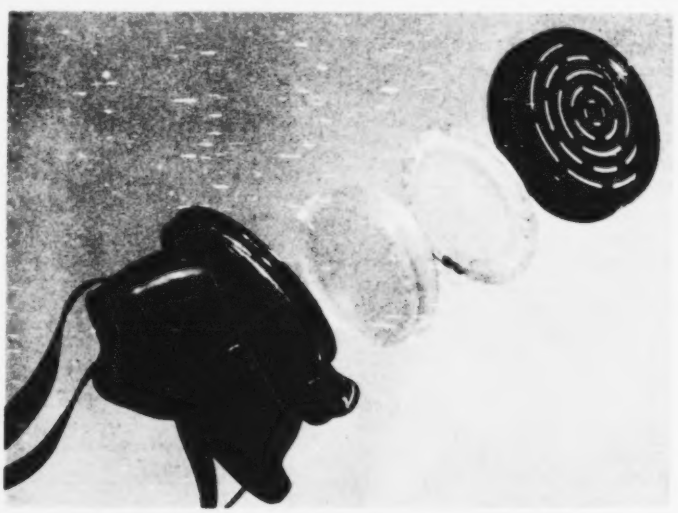


Figure 35.

Sedimentation Samplers

SETTLING PLATES Agar

General Description: The open Petri dish is probably the oldest method of air sampling and at times is still used to determine in a general way the types of micro-organisms present in the air. However, the sedimentation method of air sampling cannot be recommended when quantitative data are desired. Furthermore, open agar plates, exposed to the atmosphere, collect only those particles large enough to settle to the plate within the exposure period.

Collection Surface: Agar.

Source: Any scientific laboratory glassware supplier.

Reference: 16.

Figure 36.

SURFACE SAMPLERS

General Description: A small patch of cloth or paper can be used as a sampler when it is necessary to determine the number or type of micro-organisms which collect on walls, clothing, or other surfaces.

The patches are fastened to the surface, prior to the settling of the micro-organisms, by means of pins, adhesive, or other suitable means. If practical, sampling patches made of the same material as the sampled surface should be used. Cotton swabs can be used for sampling any solid surface.

After the samplers have been exposed for the required time, they are placed in bottles of sterile liquid, shaken for at least 5 minutes, and the liquid is assayed according to standard bacteriological techniques.

Optimum Flow Rate: None. Sampling depends upon particles settling out of the air onto exposed patch samplers.

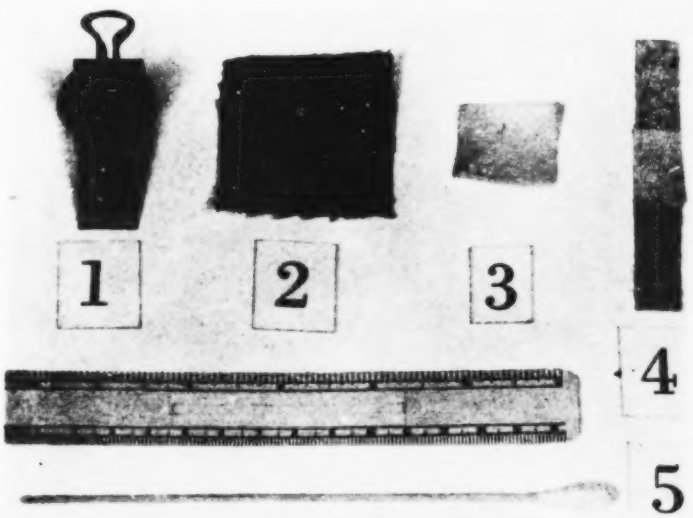


Figure 37.
(1) metal; (2) cloth; (3) paper; (4) cloth on tape; (5) cotton swab.

Collection Surface: Metal, cloth, or filter paper.

Source: Filter paper (Glass Fibre Filter Web) Catalogue Nos. CT 75428 and CT26066: Mine Safety Appliance Co., 201 North Braddock Avenue, Pittsburgh 8, Pa

GRAB SAMPLER

General Description: Any container in which a sample of air can be trapped for subsequent assay may be used as a grab sampler. The sampler design and methods of sampling vary, depending on the results desired (determination of total organisms or particles and particle size). An evacuated flask of known volume may be used to collect an air sample. The collected organisms are subsequently removed by washing with a sterile liquid. Air samples can also be trapped in a chamber that is designed so that all particles settle on a glass slide or membrane filter for subsequent size measurement, or on an agar plate for viable particle determination.

Source: No commercial source. Drawings available from Technical Development Laboratories, Communicable Disease Center, U.S. Public Health Service, P.O. Box 769, Savannah, Ga.

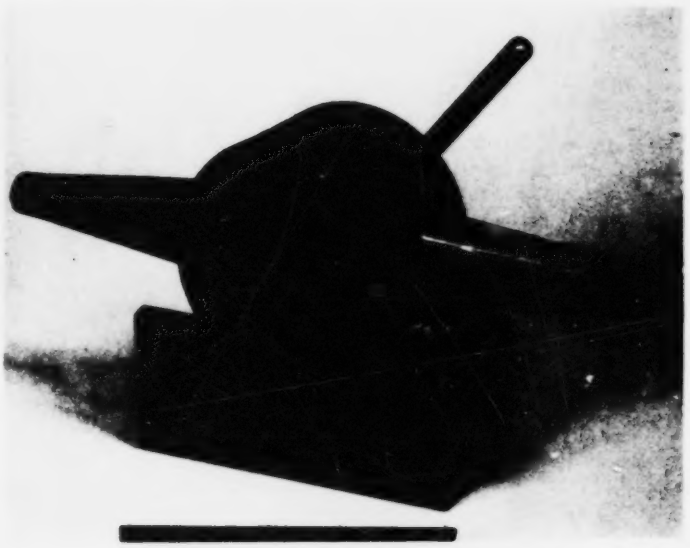


Figure 38.

MEMBRANE FILTERS

General Description: Membrane filters placed in plastic dishes may be used as fallout samplers in the same manner as an agar settling plate. The dish containing the filter and absorbent pad is placed on a flat surface and exposed to the air. After collection of the fallout sample, a broth medium is added to the pad and the sample is incubated in accordance with standard bacteriological methods.

See Figure 20 page 30

Optimum Flow Rate: None. Sampling depends upon particles settling out of the air onto exposed filters.

Collection Surface: Membrane filter.

Sources: Millipore Filter Corp., Watertown 72, Mass. Carl Schleicher & Schuell Co., Keene, N.H.

References: 23, 24, and 25.

Centrifugal Samplers

WELLS AIR CENTRIFUGE

General Description: The basic component of the Wells air centrifuge is a rotating glass cylinder. Thirty-five ml. of collecting fluid or warm melted agar are added to this cylinder, which is then placed vertically inside the sampler. The inlet tube of the sampler extends well down into the glass cylinder when it is in position. The cylinder is rotated by a motor at speeds up to 3,500 rpm. The fluid or agar covers the inside of the cylinder after a few seconds of rotation. A blower draws air through the apparatus and suspended particles are driven outward onto the inner wall of the cylinder by centrifugal force. Airflow is controlled by the speed of rotation or by the size of the inlet orifice or venturi. If a liquid collecting fluid is used, it is subsequently diluted and plated as with any liquid type sampler. If agar is used, the tube is incubated and colony counts are made through the glass wall.

Optimum Flow Rate: 28.42 liters per minute. Rate dependent upon speed of rotation and size of venturi nozzle orifice.

Collection Surface: Liquid or agar.

Source: No commercial source.

References: 16, 42, and 43.

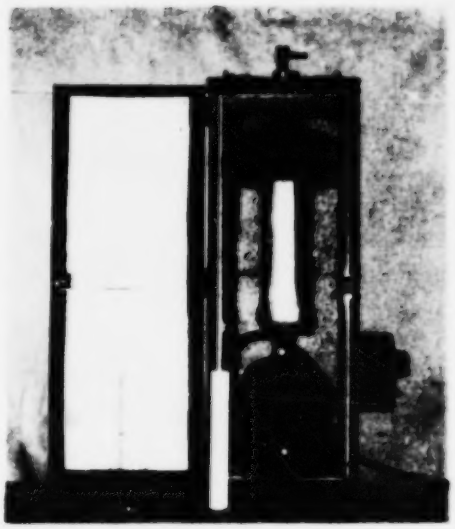


Figure 39.

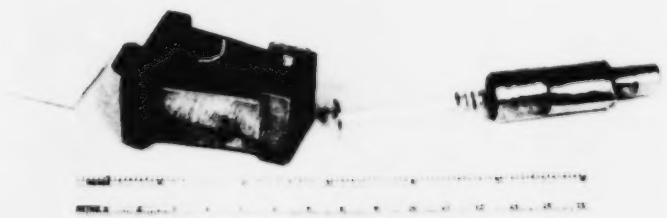


Figure 40.

AEROTEC TUBE

General Description: This is a cyclonic type sampler in which sampled air is drawn through two slits located on opposite sides of the sampler. It is designed so that the incoming air flows in a rapid spiral pattern. The larger particles drop out in the settling chambers and are collected in an attached glass vial or on a glass slide. The smaller particles pass through the cyclone section and are collected on a membrane filter. The size at which separation takes place can be changed by varying the airflow. The number of viable organisms associated with the larger particle-size group is determined by standard diluting and plating procedures. Smaller particles are analyzed by the same method used for membrane filters (p. 30). By direct microscopic examination of the slide and membrane it is possible to measure the size of the collected particles.

Optimum Flow Rate: 28.3 liters per minute.

Collection Surface: Glass.

Source: The Aerotec Corp., Greenwich, Conn.

Reference: 82.

CONIFUGE

General Description: This sampler is essentially an air centrifuge formed by two concentric cones which are separated by a narrow annular space through which air is drawn. Particles suspended in the air are impacted on the inner surface of the outer cone according to their size. The larger particles are collected at the top of the cone and the smaller particles are deposited farther down. A glass microscope slide or membrane filter covering a slit in the outer cone is used to collect a sample of the impacted particles. The number and size of the collected particles, determined microscopically, are used to calculate the size distribution of the aerosol.

Optimum Flow Rate: 25 ml. per minute.

Source: C. F. Casella and Co., Ltd., Regent House, Fitzroy Square, London, England.

References: 44.

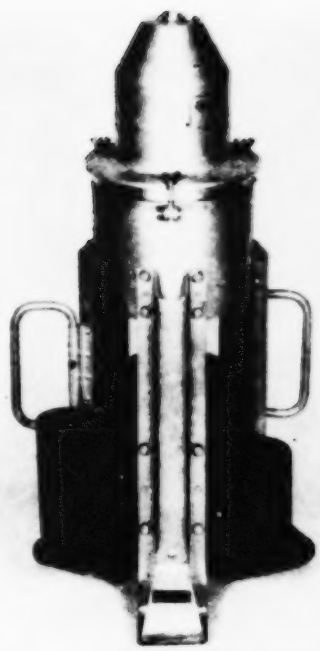


Figure 41.

ELECTROSTATIC PRECIPITATOR

General Description: The electrostatic sampler, a self-contained unit, requires an electrical potential of 7,000 volts across 2 pairs of electrodes between each of which a Petri dish is inserted. The bottom electrode, on which the agar plates rest, is charged positively in one pair, negatively in the other. Incoming air passes at the rate of 14.15 liters per minute through each hole in the center of the top electrodes. Positively charged airborne micro-organisms are deposited on the agar plate resting on the negatively charged electrode and negatively charged cells are deposited on the positively charged side. Agar plates are placed on each electrode, the cover is snapped into place, the cord inserted into a standard 115v., a.c. outlet, and unit is operated for whatever period is desired.

Although 28.3 liters per minute of air pass through the sampler, only 14.15 liters per minute pass over each charged plate. Therefore, the sum of the colonies on the two Petri dishes represents the total number of bacteria in 14.15 liters of air in a 1-minute sampling time.

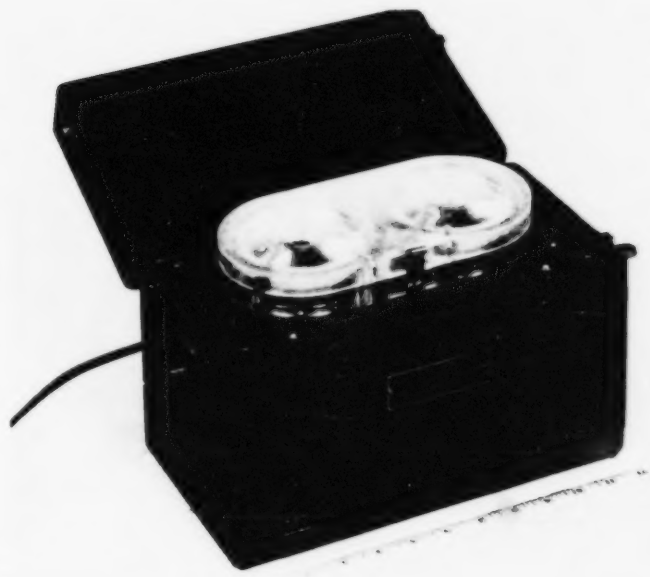


Figure 42.

Optimum Flow Rate: 28.3 liters per minute.

Collection Surface: Agar.

Source: General Electric Co., Lamp Department, Nela Park, Cleveland 12, Ohio.

References: 47, 48, and 49.

THERMAL PRECIPITATOR, WATER COOLED

General Description: This sampler operates on the principle that when an aerosol flows between a hot and cold surface the suspended particles precipitate upon the cold surface. The lower portion of this sampler is kept cool by running water. The upper portion is heated by an electrical element. The aerosol enters the sampler through the central tube and flows radially outward between the heated and cooled surfaces and out through the exhaust tube. When collecting bacteria and liquid aerosols, the temperature gradient is limited to less than 80° C. per 0.25 cm. to prevent thermal damage and evaporation of the collected material.

Optimum Flow Rate: 0.4 liter per minute.

Collection Surface: Glass.

Source: Roy A. Martin Co., 899 Wellesley Drive N.W., Atlanta 5, Ga.

References: 50, 51, and 81.

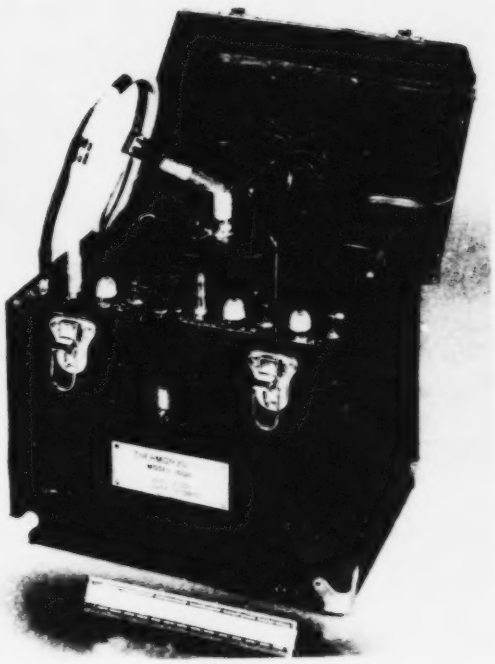


Figure 43.

THERMAL PRECIPITATOR, HOT WIRE

General Description: This precipitator, designed specifically for particle sizing, uses a hot wire to repel the particles and deposit them on a microscope glass coverslip. The collection efficiency is very high for particles of less than 5 microns in diameter, especially those in the submicron range. An oscillating model moves the collection surface so that the particles are not collected in a line, but are evenly spread over the surface. Particles may be collected on a coated grid for electron microscope study.

Optimum Flow Rate: 7 ml. per minute.

Collection Surface: Glass.

Source: C. F. Casella and Co., Ltd., Regent House, Fitzroy Square, London, England. Mine Safety Appliance is U.S. outlet for Casella Co.

References: 50, 51, and 81.

Figure 44.

MEMBRANE FILTER HOLDERS

Purpose: To hold filter media.

General Description: Several holders have been designed to support membrane filters and other sheet fiber materials for sampling airborne particles. Most holders consist of a cylindrical body, one end of which is adapted for connection to a vacuum source. The other end is fitted with a circular supporting screen or carbon pad. A clamping device seals the filter between the supporting screen and an inlet cup to prevent bypassing of the sampled aerosol. When the inlet cup is fitted with an adapter, samples may be taken in a closed system or from an air duct. One type of holder (fig. 45) is designed to accommodate both the unringed and the ringed membrane filter (filter mounted on a plastic ring). The plastic ring holds the filter against the supporting screen and eliminates the need for an inlet cup. The materials of construction and sealing devices vary with designs.

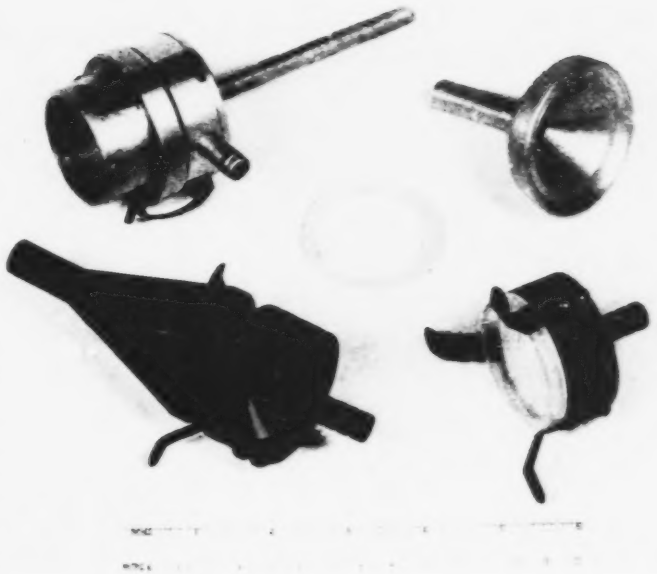


Figure 45.

Sources: Millipore Filter Corp., 36 Pleasant Street, Watertown 72, Mass. Gelman Instrument Co., Chelsea, Mich. Drawings of the membrane filter holder (fig. 45) available from Technical Development Laboratories, Communicable Disease Center, U.S. Public Health Service, P.O. Box 769, Savannah, Ga.

References: 76.

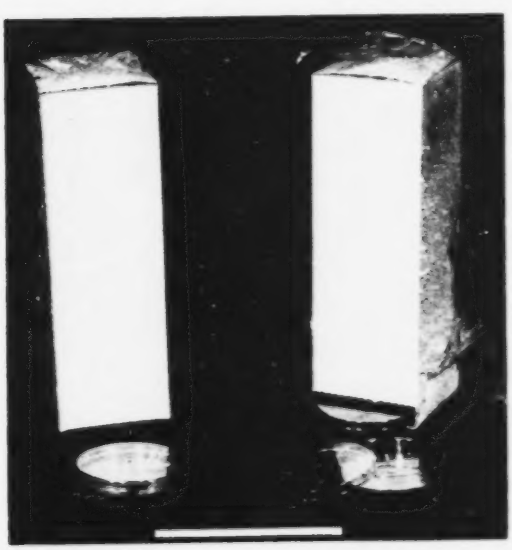


Figure 46.

DISPOSABLE PLASTIC STERILE PETRI DISH

Purpose: For bacteriological assay.

General Description: These dishes are made of clear polystyrene plastic and are commercially available in various sizes for use with such samplers as membrane filters and slit samplers.

Source: Falcon Plastic Products, 6020 West Washington Boulevard, Culver City, Calif.

References: Manufacturer's catalogue.

FLOWMETER Orifice Type

Purpose: Measurement of sampling rate.

General Description: This simple flowmeter consists of a restricting orifice, through which the air flows, and a manometer to measure the pressure drop across the orifice. After calibration, the pressure drop indicates the flow. Although orifice flowmeters are commercially available, a simple flowmeter can be constructed in the laboratory for use when extreme accuracy is not required. A short length of capillary tubing is inserted in the vacuum line of the sampler between two T-connections to which a manometer is attached. The manometer may be a commercial type, or a glass or plastic U-tube half filled with water, mercury, or other suitable fluid. The meter can be calibrated by attaching a standard flowmeter to the inlet of the sampler and recording the manometer reading corresponding to each flow indicated on the standard meter.

Sources: Fisher Scientific Co., 7722 Woodbury Drive, Silver Spring, Md. Central Scientific Co., 1700 Irving Park Road, Chicago 13, Ill.

References: Manufacturers' catalogues.

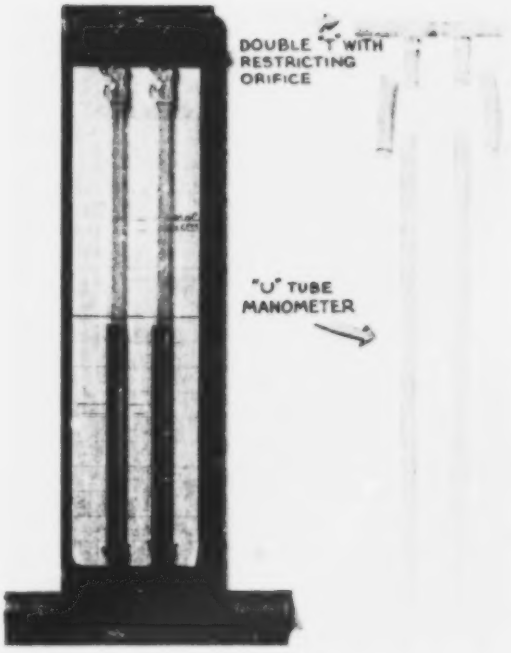


Figure 47.

FLOWMETERS Variable Area Type

Purpose: To measure rate of airflow in air sampling devices or laboratory equipment.

General Description: These variable area flowmeters consist of a float in a tapered tube mounted vertically with the small end of the taper in the downward position. The air flows into the small end, upward within the tube around the float, and leaves at the larger upper end. As airflow increases, the upward fluid pressure plus the fluid buoyant effect exceeds the downward pressure exerted by the weight of the float. This increase in pressure causes the float to rise to a point where the upward and downward forces are equalized. Weight, size, and materials of construction of flowmeters vary according to type and manufacturer.

Optimum Flow Rate: Dependent upon model selected. Available in sizes from 50 ml. per minute to 396 liters per minute.

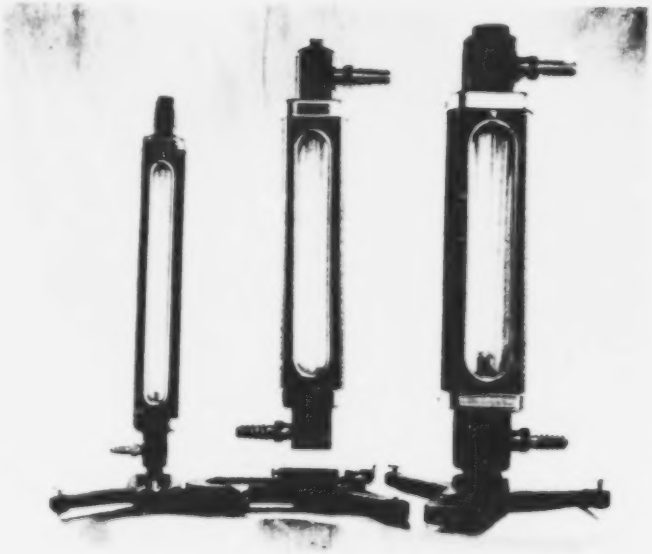


Figure 48.

Sources: Brooks Rotometer Co., Lansdale, Pa. F. W. Dwyer Manufacturing Co., P.O. Box 373, Woodland at Greenwood, Michigan City, Ind. Fischer and Porter Co., 51 County Line Road, Hatboro 6, Pa.

References: Theory of operation and limitations are found in the manufacturers' catalogues.

FLOWMETER (Wet Test Gas Meter

Purpose: To measure and calibrate airflow of sampling equipment.

General Description: Gas is forced into one of four chambers. The buoyancy of the gas causes the chamber to rise in the water, resulting in the rotation of the rotor and release of the gas from the meter as the chamber reaches the top. This procedure is repeated as long as gas flows through the meter and the consecutive compartments are filled with gas at each quarter turn of the rotor.

Optimum Flow Rate: Dependent upon model selected. Available in sizes from 566 liters per hour (20 cubic feet per hour) to 4,240 liters per hour (150 cubic feet per hour).

Sources: Precision Scientific Co., 3737 West Cortland Street, Chicago 47, Ill. American Meter Co., Inc., 991 Broadway, Albany 1, N.Y.

Reference: Theory of operation and limitations are found in manufacturers' catalogues.

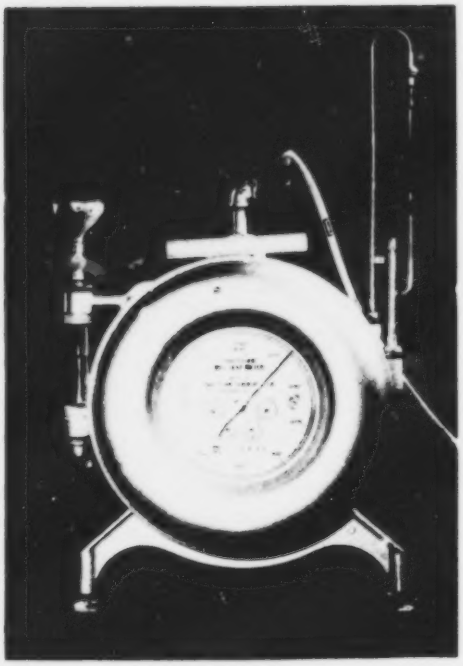


Figure 49.

VACUUM PUMP

Purpose: To provide vacuum for sampling.

General Description: Vacuum pumps suitable for sampling are commercially available in several types and sizes. They may be of the rotary vane, diaphragm, or centrifugal type and may be powered by electric or gasoline motors. Some can be operated by hand. The pump selected will depend on sampling requirements, availability, and so on. Decision on which pump to select must be left to the investigator. A convenient pump for operating most samplers is the electric Gast 1/6 hp., 1,750 rpm vacuum pump which is compact and will maintain a critical flow of approximately 15 liters per minute.

Sources: Leiman Brothers, Inc., 146–181 Christie Street, Newark, N.J. Gast Manufacturing Co., Benton Harbor, Mich.

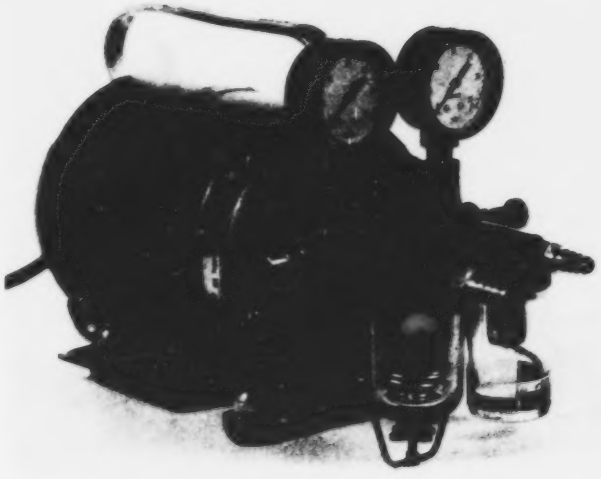


Figure 50.

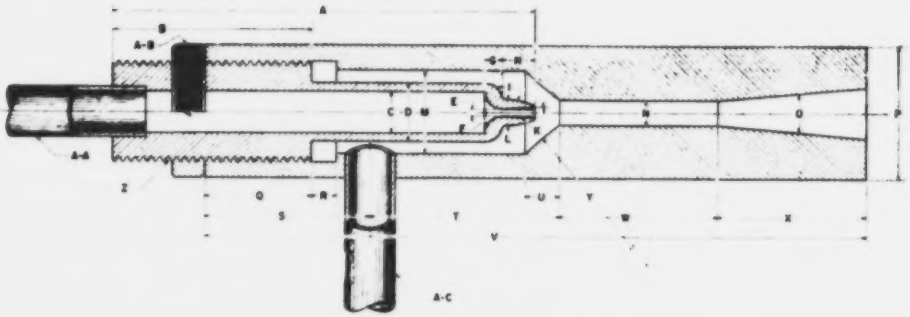


TABLE OF DIMENSIONS

	SMALL	MEDIUM	LARGE		SMALL	MEDIUM	LARGE
A	2 1/4"	2 3/4"	3 3/16"	P	1/2" Dia	3/4" Dia	1" Dia
B	1"	1 1/2"	2"	Q	3/4"	3/4"	3/4"
C	1/8" Dia	7/32" Dia	5/16" Dia	R	1/8"	5/16"	1/4"
D	5/16" Dia	5/16" Dia	7/16" Dia	S	1 1/8"	1 1/8"	1 1/4"
E	.045" Dia	.043" Dia	.083" Dia	T	7/8"	1"	1 1/4"
F	WELL ROUNDED			U	1/4"	5/16"	3/8"
G	1/8"	3/16"	1/4"	V	3 1/4"	3 7/8"	4 7/8"
H	1/8"	3/16"	1/4"	W	5/8"	1"	1 3/16"
I	WELL ROUNDED			X	1/2"	3/4"	1 1/8"
J	.045" Dia	.043" Dia	.084" Dia	Y	1" R	1 1/2" R	2 R
K	STRAIGHT WALLED	STRAIGHT WALLED	TAPERED WALLED	Z	5/16"-24 NF THREAD	5/16"-18 NF THREAD	5/16"-18 NF THREAD
L	.06" Dia	12" Dia	20" Dia	A-A	1/4" O.D.	5/16" O.D.	3/8" O.D.
M	1/4" Dia	1/2" Dia	5/8" Dia	A-B	1/8" Dia STOP NUT	3/16" Dia STOP NUT	1" Dia STOP NUT
N	.075 Dia	1/8" Dia	3/16" Dia	A-C	1/4" O.D.	5/16" O.D.	3/8" O.D.
O	10°	10°	10°				

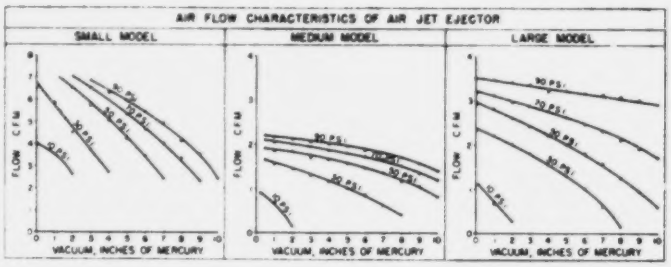


Figure 51.

VACUUM PUMP (Jet Type)

Purpose: To provide vacuum for sampling.

General Description: These vacuum pumps are ejectors or aspirators. They are operated by converting the pressure head of some fluid into a high velocity stream which produces a vacuum in the suction chamber of the device. Several such devices are available, each with different capacities, and they are designed to use compressed gases or water under pressure for the operating medium. Most of the devices designed specifically for aerosol sampling are of moderate capacity, and a few are available in complete sampling kits.

Sources: Union Industrial Equipment Corp., 20 Davis Avenue, White Plains, N.Y. Schutte and Koerting, Cornwells Heights, Pa. Penberthy Injector Co., Division of Buffalo-Eclipse Corp., 1242 Holden Avenue, Detroit 2, Mich.

AUTOMOBILE ENGINE AS VACUUM SOURCE

Purpose: To provide vacuum for sampling.

General Description: A convenient available source of a sampler vacuum is the intake manifold of an automobile engine. Samplers can be connected to the intake manifold of an engine either by means of the windshield wiper connection or a separate specially installed connection. These samplers can be operated at flows up to 82 liters per minute. The engine should be run at idling speed. This allows operation for hours without overheating.

Reference: 75.

Table 9. Airflow through two sampling devices utilizing automotive engines as vacuum source

Vehicle type	Engine speed	Airflow (cfm) through	
		Sieve sampler [1]	Tangential jet impinger [2]
International, K3, 1-ton truck, 1945, 16,964 miles.	Idle	3.5	2.1
	Medium	3.5	2.1
	High	(3)	(3)
International, ½-ton truck, 1947, 16,574 miles.	Idle	3.3	1.8
	Medium	3.5	2.0
	High	(3)	(3)
Jeep, U.S. Army, 1942, 25,452 miles.	Idle	3.4	2.2
	Medium	3.5	2.1
	High	3.4	(3)
Jeep, Willys, Universal, 1948, 21,980 miles.	Idle	2.9	2.0
	Medium	3.0	1.9
	High	3.0	(3)
Chevrolet, Model 3100, Carryall, 1,100 miles.	Idle	3.0	2.3
	Medium	3.0	2.3
	High	2.8	(3)
Ford, 6 cylinder, 1949, 21,980 miles.	Idle	3.1	2.1
	Medium	3.1	2.3
	High	(3)	(3)
Ford, 8 cylinder, 1948, 37,120 miles.	Idle	1.9	2.0
	Medium	2.9	2.1
	High	2.0	(3)
Buick, Special, 1948, 45,788 miles.	Idle	2.8	1.7
	Medium	3.0	2.0
	High	(3)	(3)

[1] Sieve sampler orifice: 340 holes, 0.0145-inch diameter.
[2] Tangential jet impinger orifice: 1 hole, 0.0625-inch diameter.
[3] Engine overheated.

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